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# THE DEVELOPMENT AND PRESENT PROBLEMS OF SOIL MICROBIOLOGY

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## 1. Development of soil microbiology

IT is necessary when reviewing soil microbiology to attempt to define the nature and scope of the subject. The soil is that region on the earth's surface where geology and biology meet. Geological materials from below and organic remains from above are the parent materials from which soil is formed mainly by the activities of micro-organisms. These micro-organisms are responsible for turning the raw materials into soil; they affect the character of the soil that is formed and, within the soil, they influence the supply and uptake of nutrients by plants growing therein and also their condition of health or disease. We may therefore define soil microbiology as being a science aimed at discovering how particular micro-organisms behave in the soil and what are the effects of this behaviour on the nature of the soil and on the growth of higher plants therein. It appears that there is no environment on earth where so great a variety of micro-organisms can be found in so small a space, as in the soil. From it one can isolate organisms of great academic interest because of their morphological or biochemical peculiarities which have been studied for reasons quite other than their likely importance in the soil. Soil is constantly combed for new organisms that produce antibiotics of possible use in medicine. Such investigations, important though they may be in other connexions, should not be included under soil microbiology just because the organisms concerned happen to have come from the soil. Again, soil chemistry is constantly concerned with changes that are brought about by micro-organisms, but the study of such changes should only be called soil microbiology when the particular organisms that bring about these changes are being studied.

Soil microbiology came rather late in the field and, when it began, soil chemists had already identified the major plant nutrients in the soil. They had also obtained evidence that the oxidation of ammonia to nitrate and the fixation of atmospheric nitrogen were biological processes. The first big advances in soil microbiology, which took place in the last decade of the nineteenth century, were due to the development by Winogradsky[1] and by Beijerinck of the technique of enrichment culture. In this technique, a soil inoculum is added to a selective medium designed to encourage the differential growth of the organisms capable of carrying out the chemical changes that are being studied. By serial transfer of the resulting culture into fresh media, a growth of organisms can be obtained in which the desired organisms predominate, and these are then isolated by plating or dilution in suitable media. By this technique many important soil organisms were isolated and described and the microbiological basis of a number of important chemical transformations was established. Nitrogen fixation, ammonification, ammonia oxidation, nitrate reduction, sulphur oxidation, cellulose decomposition and so forth were thus recognized as due to microbial action.

The dramatic success of this technique, however, had its disadvantages. In the first place it led to the attribution of particular chemical changes known to take place in the soil to particular organisms that happened to have been isolated from enrichment cultures, usually with no proof that these organisms were responsible for such changes. In the second place it led to the classification of organisms into separate ' working parties ' such as ammonifiers, denitrifiers, and so forth, in defiance of the obvious fact that most organisms can carry out different processes according to the nature of their environment. Indeed most soil organisms, living as they do in a very heterogenous environment, have developed a correspondingly varied system of enzymes, enabling them to cope with a great variety of substrates. And lastly, the subdivision of the soil microflora into clear-cut groups tended to obscure what is the main problem of the soil microbiologist —emphasized by Winogradsky himself—namely the complex interactions that take place between the components of soil population. It was the discovery by Russell & Hutchinson[2] that treatment of the soil with a mild antiseptic could increase microbial activity, that drew particular

attention to the equilibrium that exists within the soil population between different competing groups, by showing that this equilibrium could be disturbed by a treatment that eliminated certain groups.

## 2. Components of the soil micropopulation

This realization of the complexity of the soil population has led to extensive surveys of the main groups of organisms and to attempts to develop methods for enumerating them. On the qualitative side, surveys of the most common taxonomic groups of soil bacteria have been made. These have emphasized the predominance of organisms resembling the Corynebacteria (Clark,[3] Topping[4] and Jensen[5]) and of Gram-negative non-sporing rods attributable to the Achromobacteriaceae and the Pseudomonadaceae (Lochhead & Chase[6]), and the great variety of spore-forming bacteria (Smith, Jordan & Clark[7]) in soil. A different type of classification has been made by Lochhead and his school (Lochhead & Chase ;[6] Stevenson & Rouatt[8]). This classification is based on the nutritional requirements of the bacteria which are grouped according to whether they are able to grow on a simple medium, or require certain amino-acids, growth substances, yeast extract or soil extract. Soils have been found to differ markedly in the proportion of the different nutritional groups found in their bacterial populations. This presumably reflects differences in the composition of the organic matter of the soils themselves.

That interesting group, the Myxobacteria, is well represented in soil and has been studied by Singh[9] and by Noren.[10–13]

Qualitative surveys of the common soil fungi (Chesters,[14] Warcup[15] and Garrett[16]) and actinomycetes have also been made, and a considerable population of algae (Lund [17–19]) has been found in some soils. Algae are not limited to the surface but also occur within the soil where they must live heterotrophically.

The soil also contains an abundant population of protozoa of which flagellates and amœbae (Singh[20]) are the most abundant groups, as well as Myxomycetes (Singh[21, 22]). The importance of these groups lies in the fact that they feed on soil bacteria and are selective in their bacterial food (Singh[23]).

On the quantitative side it cannot be said that we have yet devised altogether satisfactory counting methods. In the case of bacteria, actinomycetes and fungi, colony counts from platings of a diluted soil suspension have been the standard method. Even with bacteria this method meets with the difficulty that no one medium is suitable for all the nutritionally varied organisms present. It also involves a further quandary. Recent work, especially that of Lochhead and his fellow workers, referred to above, has shown that many soil bacteria require quite a complex medium including growth substances. On the other hand, if the medium used for plating is too rich, competition between organisms on the plate will reduce the colony count. Such difficulties have instigated the development of direct counting methods in which microscope counts of the organisms are made from stained films of soil suspension (Jones & Mollison[24]). These methods of course suffer from the disadvantage that the organisms are killed in making the preparation and cannot be isolated and studied, nor can they usually be identified by shape and size, since organisms in soil may develop a morphology different from that shown in culture. Direct microscope counts, however, give far higher numbers of bacteria than do plate counts. The former may reach several thousand million per gramme of soil where the plate count from the same sample may give estimates of 50 to 100 millions (Skinner, Jones & Mollison[25]). The cause of this large discrepancy is not yet clear. It is very important that it should be ascertained since if it is due to the differential nature of the plating media used, it means that the majority of bacteria in the soil have so far escaped cultivation and study. It is likely that the higher estimate derived from microscope counts more nearly represents the actual numbers of bacteria. Jensen[26] compared the amounts of $CO_2$ evolved from incubated samples of various soils with the bacterial numbers estimated by both methods and found that estimates derived from microscope counts were correlated with the amount of $CO_2$ evolved and were of the order that might be expected from the biochemical activity of the soils. With regard to fungi, plate counts suffer from the additional uncertainty as to whether a colony is derived from a spore or from a fragment of mycelium of uncertain size. With the microscope count, fragments of hyphae can be counted

and measured and the result expressed as meters of mycelium per gramme, a figure that has at least some apparent meaning. No really satisfactory method for counting actinomycetes in soil has yet been developed. Microscope counts of ' bacterial cells ' probably include actinomycete spores.

There is a number of soil organisms, such as protozoa and certain bacteria, that will not produce colonies on plates and are not sufficiently numerous in soil to be counted directly. For such organisms a dilution method has to be used (Singh[27]).

These various quantitative methods, while they leave one uncertain as to the absolute numbers of organisms in soil, have a value in comparing estimates made by the same technique from a number of soil samples. Estimates made either by plating or by microscope counts from replicate subsamples of a sifted and well mixed soil sample, agree within random sampling expectation. It has thus been possible, for example, to demonstrate differences in the numbers of micro-organisms in differently manured plots, differences correlated with depth in the soil profile, and fluctuations in numbers in the surface soil from the same plot in samples taken at different times.

### 3. Localized distribution of the soil micropopulation

In such quantitative work, however, each sample of soil has first been well mixed and then a well-shaken suspension of the soil has been used for making the count. The sample has in fact been homogenized. This tends to obscure the fact that in the soil sample, as taken from the field, the micro-organisms will not in fact be distributed uniformly. It seems likely that they occur most abundantly in and around particles of organic matter and in the colloidal material surrounding the mineral particles. A really satisfactory method for observing directly the organisms in the soil itself is still required. Alexander & Jackson[28] have recently developed a method in which a column of soil taken from the field is treated with a stain differential for micro-organisms, impregnated with plastic and then sectioned by the method used for rock specimens. Such sections enable one to observe the distribution of the colloid film surrounding the mineral particles and some of the micro-organisms lying in it. This is the first method devised which gives a direct impression of the localization of micro-organisms in the soil. Some very interesting indirect evidence, that at least some soil organisms grow and exert their activities when attached to the soil colloids, has been obtained by Lees & Quastel[29–31] in their biochemical study of the oxidation of ammonia in soil. They found that the rate of nitrification of a given quantity of ammonium sulphate in soil was proportional to the degree to which the ammonium ions were absorbed on or combined in the base-exchange complex of the soil and that, in consequence, the addition of sterile soil to a nitrifying soil will increase its rate of nitrification in proportion to the base-exchange capacity of the added soil.

A particular example of the uneven distribution of bacteria in the soil is that shown on the surface and in the immediate neighbourhood of the roots of living plants, an environment that has come to be known as the ' rhizosphere '. In spite of considerable sampling difficulties, it has now become clear that the micro-organisms are much more numerous in this region than in the main mass of the soil away from the roots (Katznelson, Lochhead & Timonin;[32] Clarke[33]). It has also been found that the types of bacteria predominant in the rhizosphere are different from those away from the roots, particularly as regards those that are dependent on amino-acids for their nutrition (Lochhead & Thexton;[34] Wallace & Lochhead[35]). It seems likely that these differences are due largely to substances secreted by the roots, but we need much more information as to what these substances are. A number of these is listed by Winter[36] and Harley.[37] Timonin[38] was able to associate rhizosphere differences between varieties of flax with HCN secretion.

The rhizosphere population must have an important effect on the nutrition of the plant since compounds in solution must pass through it to reach the roots. Moreover, insoluble compounds may be brought into solution by acids produced by the rhizosphere microflora. Gerretsen[39] showed that micro-organisms capable of dissolving insoluble phosphates occur on plant root surfaces. Growth-stimulating substances such as $\beta$-indolylacetic acid are known to be produced by a number of soil organisms, e.g. *Rhizobium*, and these may well have a direct effect on root growth. Antibiotic production by the rhizosphere organisms may also have an importance in

checking the attack of root-pathogenic fungi. On the other hand, substances toxic to the root may be produced by rhizosphere micro-organisms. Steinberg[40] produced symptoms of toxicity in tobacco with pure cultures of various bacteria, including species not usually regarded as pathogenic, which he supplied to the root surroundings. Thus the study of rhizosphere micro-organisms is a field of work now being actively pursued which is likely to produce important results both as regards the nutrition of crop plants and also in relation to root disease.

Some more general aspects of microbial activity in soil may now be considered. This activity may be grouped under three interrelated headings: (1) its influence on the formation and structure of the soil; (2) its influence on plant nutrients and (3) its effects on root disease.

## 4. Effects of microbes on soil structure

As regards soil formation it is clear that, in general, this results from microbial decay of organic remains, but the nature of the resulting soil is also influenced by microbial action. A feature of very great agricultural importance in the soil is its crumb structure, that is to say the binding together of fine soil particles to form water-stable aggregates. This problem has given rise to a great deal of research, especially because of its important bearing on soil erosion. There appear to be two types of structure improvement. In the first place, there is a slow improvement resulting from long continued supply of organic material to soil (Swaby[41]). This type of improvement is probably due to some fraction of the humic material slowly produced by microbial activity, but the slowness of the action and the chemical complexity of the humic material involved has made its elucidation too difficult up to now. Secondly, there is also a rapid improvement in soil aggregation which follows the addition to soil of some readily decomposable organic materials. Such improvement is usually temporary in character. It seems clear that it results directly from microbial action and it has been produced in sterile soil inoculated with pure cultures of certain micro-organisms. These include bacteria that secrete polysaccharide gums (Geoghegan & Brian;[42] Martin[43]) and also actinomycetes and fungi which appear to bind the particles together by mycelial growth (McCalla;[44] Martin & Anderson[45]). Swaby[46] considered that the binding action of fungal mycelium was the most important factor, and in an experiment where a number of organic substances were added to unsterilized soil samples, he found a close correlation between the degree of aggregation of the soil and the quantity of fungal mycelium estimated by microscope determination in stained films of soil suspensions. Further investigation of the role and relative importance of polysaccharides and of mycelium in forming crumbs is needed.

## 5. Effects on plant nutrients

The relation of micro-organisms to the supply of nutrients to the crop is a wide subject within which it is only possible to refer to some of the more important aspects. It has become clear from the work of soil chemists that the compounds of many of the elements that are important to plant nutrition pass through cycles of transformation in soil. It has long been realized that micro-organisms are largely concerned in the cycles of nitrogen and carbon in the soil, but the partial elucidation of the part that they play in the cycles of such other elements as iron, manganese and sulphur has been a more recent development in which soil microbiology, soil biochemistry and soil chemistry have all had their share.

### (*a*) *The manganese cycle*

Manganese has a great importance in soil, first because it is a minor but essential nutrient whose absence in an available form results in certain crop diseases such as 'grey speck' in oats and 'marsh spot' of peas, and secondly because in the form of manganese dioxide it has the power of oxidizing a number of toxic compounds formed in soil under reducing conditions. As far as is known, only divalent manganese is used by crop plants and even in this form it may be bound to organic matter in an unavailable state (Heintze & Mann[47]). When a manganese salt is added to a mineral soil of neutral or slightly alkaline reaction, oxidation may take place (Mann & Quastel[48]) to trivalent manganese which later gives rise to $MnO_2$ (Dion & Mann[49]). It can be shown that this oxidation is almost wholly due to micro-organisms since the action is stopped by heating the

soil or by adding such biological poisons as sodium azide. A number of soil micro-organisms can oxidize manganous salts (Bromfield & Skerman ;[50] Gerretsen ;[51] Leeper & Swaby ;[52] Timonin[53]) particularly in the presence of hydroxy-acids in an alkaline substrate (Sohngen[54]). It is quite likely that organisms exist that can oxidize manganous salts autotrophically, but this has not yet been clearly demonstrated. The reduction of $MnO_2$ in soil under conditions of limited oxygen supply is at least partly due to micro-organisms. A number of compounds produced by micro-organisms such as polyphenols and thiol compounds can reduce $MnO_2$, so that this process is relatively non-specific from the bacteriological point of view.

(*b*) *Oxidation and reduction of iron compounds*

The oxidation and reduction of iron salts in soil is largely brought about by organisms. A number of bacteria, algae and flagellates (Pringsheim[55, 56]) deposit ferric oxide, but it is not established in most cases whether the oxidation involved is autotrophic. One organism however, *Thiobacillus ferroxidans*, has been definitely shown to oxidize ferrous iron autotrophically, deriving its energy needs from the reaction (Temple & Colmer[57]), and there are probably others. The water organism *Gallionella* has been shown to be a true autotroph (Sartory & Meyer[58]). Bromfield[59] studied the reduction of ferric salts in soil. One of the most active organisms was *Bacillus circulans* which reduces ferric to ferrous salts in anaerobic culture, but only if supplied with a compound capable of yielding oxygen, which it cannot obtain from the ferric salt. It is probable that much of the transformation of iron salts is brought about indirectly by changes due to non-specific organisms. Thus Bloomfield[60] has found that soluble substances produced by rotting vegetation can actively reduce ferric oxide.

(*c*) *Trace elements*

A field of work that needs development is that concerning the role of micro-organisms in affecting the availability of other trace elements in soil, such as copper, zinc, cobalt and molybdenum (see review by Starkey[61]). Biochemists have shown that such minor elements are contained in important groups of enzymes. This may be their function in higher plants and it is not surprising that they are also essential to the growth of a number of micro-organisms (Mulder[62]). These requirements have been much studied of late as regards fungi (Perlman[63]) and test micro-organisms have been used to assay a number of minor elements such as manganese (Bentley, Snell & Phillips[64]), copper and molybdenum (Mulder[62, 65]). It may well be that micro-organisms themselves are responsible for much of the locking up of minor elements in the soil.

(*d*) *The sulphur cycle*

Changes in sulphur compounds brought about in soil may be important because of their effect on the availability of nutrients such as manganese, their effect on the pH of the soil and because of the production under certain conditions of toxic compounds. The relation of bacteria to the sulphur cycle in soil has been the subject of much recent research. The main stages of this cycle have been worked out and the organisms responsible have been isolated and studied. This field has recently been well reviewed by Butlin & Postgate[66, 67] who are themselves responsible for much of the work on the subject.

(*e*) *The nitrogen cycle*

It is, however, the nitrogen cycle which because of its special importance to crop nutrition, has always attracted most attention of soil microbiologists. The oxidation of ammonia to nitrate has attracted valuable research of late (see reviews by Meiklejohn ;[68] Quastel & Scholefield ;[69] Lees[70]). The steps in the oxidation of ammonia by *Nitrosomonas* have been investigated by Hofman & Lees[71] who have obtained strong evidence that hydroxylamine is the first intermediate. An important problem relating to nitrification is the cause of its occurrence in acid soils. The pH optimum for the strains of *Nitrosomonas* that have mostly been studied in culture has been found to be about 8·6 and the activity falls off rapidly on the acid side of neutrality,

with a limit at about pH 6. On the other hand, in soil as acid as pH 4, nitrification has sometimes been found to occur. This raises the unsolved question whether there are strains of *Nitrosomonas* that are acid tolerant or whether different organisms may not be responsible for nitrification in acid soils. The possible importance of heterotrophic nitrifying organisms has been claimed but on doubtful evidence. More specifically, organisms have been found that will nitrify pyruvic oxime (Jensen[72] and Quastel, Scholefield & Stevenson[73]). Oxaloacetic oxime will also undergo nitrification, but it is not known whether such oximes occur in soil.

The production of mineral nitrogen by decomposition of organic compounds in soil and the loss of this nitrogen by microbial assimilation have been extensively studied, and soil chemists have related the changes to the amount and nature of the organic materials in the soil. The great variety of micro-organisms concerned has, however, so far defied microbiological analysis.

An outstanding problem is the loss of total nitrogen that occurs especially in well-manured soils cropped with non-legumes. Much of this nitrogen seems to be lost as gas, some of it as nitrogen, some as nitrous oxide and a small part as nitric oxide. Wijler & Delwicke[74] have shown, by using $^{15}N$, that the losses as gas came from the soil nitrate. Four types of denitrifying organisms have been found: (1) the autotroph, *Thiobacillus denitrificans* (Baalsrud & Baalsrud[75]), (2) a facultative autotroph, *Micrococcus denitrificans* studied by Kluyver,[76] (3) a group of heterotrophic *Pseudomonads* (Allen & Van Niel[77]) and (4) certain heterotrophic spore-forming bacilli (Verhoeven[78]).

It seems clear that leguminous crops are mainly responsible in temperate regions for increases in soil nitrogen by fixation but that definite increases may occur in the absence of legumes. It is still not understood what organisms are mainly responsible for this non-symbiotic fixation. The numbers of *Azotobacter* cells found in most soils are too low to account for the nitrogen fixed unless one assumes an improbably high rate of cell multiplication and decay (Jensen[79]). Attention is now directed to the anaerobic nitrogen-fixing Clostridia. These are abundant and widely distributed in soil, and it is uncertain whether the low rates of fixation found in culture experiments may not be due to unsuitable media. Indeed, one may say that a more thorough study of anaerobic organisms in soil is overdue, since they may be of importance in a number of other processes such as the reduction of insoluble oxides of minor elements and possibly in the formation of toxic substances.

### (*f*) *Nodule bacteria and leguminous crops*

The nodule bacteria of legumes (Rhizobium) constitute a field that continues to attract extensive research. Much of this of late has been concerned with the behaviour of Rhizobium within the host plant (Nutman[80] and Allen & Baldwin[81]) and is hence not strictly relevant to this survey, but there have also been interesting advances in connexion with the organisms in the soil (Allen & Allen ;[82] Thornton[83]). These have recently centred round the problem of effective and ineffective strains. There are large differences between strains of Rhizobium in respect of their ability to fix N in the host plant, some strains being highly effective in this respect while, at the other end of the scale, strains occur that fail to fix nitrogen in the nodules. The latter constitute a problem especially in the case of clovers. Soils in hill pastures in Great Britain contain a high percentage of ineffective strains of clover Rhizobium (Thornton[83]). A similar case also occurs in some districts in Australia (Vincent[84]). It is thus a question whether the growth of clover in these areas may not be improved by seed inoculation to replace ineffective by effective strains. This raises the problem of competition between the relatively few bacteria that can be introduced by seed inoculation and the large numbers of wild clover nodule bacteria in the soil. Experiments have shown, first that some strains of Rhizobium produce nodules more readily than others, and secondly that acute competition occurs in the soil outside the roots between strains and that some of these strains are specifically dominant in competition with each other (Nicol & Thornton ;[85] Vincent & Waters[86]). By using such dominant strains for inoculation in the field it has been found possible to get over 60% of the nodules formed by the introduced strains (Read[87]), and there is evidence that the growth of clover in soils containing ineffective strains can be improved by seed inoculation with suitably selected effective strains (Jenkins, Vincent & Waters[88]).

### 6. Antagonism between micro-organisms in soil in relation to root disease

This case illustrates one of the most important fields of soil microbiology, the study of antagonism between the components of the soil population. This antagonism may be due to three main causes. First, organisms may compete for some limiting nutrient in the soil. Secondly, an organism may produce substances harmful to another organism. A special case of this is antibiotic production. Thirdly, in the case of protozoa, one organism may actually ingest another. This problem of competition attracts special attention now, because of the possibility of controlling root disease fungi either by introducing into the soil organisms antagonistic to the pathogen (Brian ;[89] Weindling, Katznelson & Beale[90]), or by encouraging the growth of those already present therein.

It has been found difficult to assess the relative importance of nutrient competition and antibiotic action between an organism in soil and its antagonist. Even where the antagonist is known to produce an antibiotic, it is by no means certain that antagonism, when found in soil, is in fact due to the antibiotic (Siminoff & Gottlieb[91]). In the case of root pathogenic fungi, antagonistic actinomycetes are so prominent on laboratory media that special attention has been given to them (Thornton & Skinner[92]). An extremist view has been put forward that actinomycete antibiotics are not operative in soil, largely because they are very liable to adsorption on to clay colloids or destruction by other organisms. Recently, Stevenson[93] working with a number of actinomycetes antagonistic to Helminthosporium has shown that specific effects on germinating spores of the fungus found *in vitro* were also reproduced if these spores were placed in soil containing the actinomycete, and that one of the actinomycetes, *Streptomyces antibioticus*, and the antibiotic actinomycin which it produces were both able to induce the same characteristic deformation of germinating hyphae in sterilized soil as well as *in vitro*. Thus in sterilized soil, some of the antagonistic effects of these actinomycetes must be due to the antibiotic action. There is also evidence that antibiotic substances of unknown origin do accumulate in soil, since germination of fungal spores is largely inhibited in fresh but not in sterilized soil (Dobbs & Hinson[94]).

### 7. The problem of altering the soil population

It has been found possible to obtain some control of fungal root disease in pot experiments with sterilized soil by inoculation with antagonistic organisms (Ledingham, Sallans & Simmonds[95]), but the problem of achieving this in unsterilized soil is much more difficult because here there is the problem of establishing the antagonist in competition with the other members of the soil micropopulation (Katznelson[96]). The introduction of a micro-organism into fresh soil by inoculation has always proved difficult, although it has been successful in the case of legume inoculation. Here it is likely that success is due to the excretion by the plant roots of substances differentially favourable to Rhizobium in the rhizosphere. It would seem that to establish an organism in soil it is necessary to render the environment differentially favourable to it. Work on the decomposition of chlorophenoxyacetic weed-killers in soil has shown that the addition of as little as 2·5 mg. of 2 : 4-dichlorophenoxyacetic acid (2,4-D) per lb. of soil will so alter the micropopulation that subsequent additions of the compound disappear at an enhanced rate (Newman & Thomas[97]). From such ' adapted ' soils, organisms have been isolated that will attack 2,4-D (Audus & Symonds ;[98] Jensen & Petersen ;[99] and Stapp & Spicher[100]). 2,4-D is an unnatural compound not normally found in soil and hence its addition provides an environment specifically favourable to the few species of organisms capable of attacking it. This case is thus a relatively simple one from the microbiological point of view and its further study may help to solve the problem of introducing organisms into soil. The solution of this problem underlies the use of antagonistic organisms to combat root disease. Recent work on systemic antibiotics that can be taken up by the roots of plants (Brian *et al.*[101]) suggests the possibility that if the problem can be solved, even diseases of the top may be controllable by establishing antibiotic organisms in the soil or rhizosphere.

There is another way in which the equilibrium of the soil population can be disturbed. This is by partial sterilization of the soil by antiseptics or heat. This has long been a practice in glasshouse soils, but recent field experiments have shown that here also such treatment results in

marked quantitative and qualitative differences being set up in the soil micropopulation that can be detected a long time after the treatment (Singh & Crump ;[102] Mollison ;[103] and Warcup[104]). The ecological factors involved require further study. Indeed the problems of soil 'micro-ecology' are fundamental to almost any practical advances in soil microbiology.

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# KAFFIRCORN MALTING AND BREWING STUDIES. I.—The Kaffir Beer Brewing Industry in South Africa

By H. M. SCHWARTZ

The brewing of kaffir beer, a traditional drink of the Bantu, is becoming an important industry in South Africa. The raw materials and processes used are described, and some of the problems associated with the change over from small-scale brewing according to the traditional Bantu method to large-scale production are discussed.

## Introduction

Kaffir beer (Zulu : *utshwala* ; Sesuto : *joala* ; Eastern Cape tribes : *utywala*) is a traditional drink of the Bantu people of Southern Africa. A number of different kinds of native beer have been described by Juritz[1] and Turner.[2] The best known is that prepared from kaffircorn (*Sorghum caffrorum* Beauv.) and other cereals, and this is what is generally meant by the term ' kaffir beer '. According to Hellman,[3] *utywala* is a generic name for beer of all kinds, while kaffircorn beer is called *mqombothi*. In this and succeeding papers in this series ' kaffir beer ' will be understood to mean *mqombothi*. It is a beverage with a pleasantly sour taste and the consistency of a thin gruel. Its alcohol content ranges from about 1 to 8% by volume,[1] although a freshly prepared beer rarely contains more than 4%.[4] The traditional methods for preparing kaffir beer in the native reserves have been described by several authors.[1, 2, 5, 6] Some of these descriptions, however, appear to be inaccurate. A good account of the method used by the Zulus in Natal has been given by Doidge.[4]

Kaffir beer is brewed by employers of Bantu labour (especially in the mines) in the Union of South Africa and the Central African Federation, and is supplied to the workers as part of their rations, since it is regarded as having a high nutritive value, particularly as a source of vitamins of the B-group.[5] It is also brewed on a large scale by many urban local authorities in the Union, who sell it at 3*d.* per pint and use the profits to provide welfare services and housing for the Bantu. The brewing of kaffir beer by municipalities in Natal dates back to about 1916,[7] whilst in the Transvaal municipal brewing was started in 1938. Municipalities in the Cape Province and the Orange Free State do not generally brew beer at the present time.

The production of kaffir beer by local authorities and private companies in the Union of South Africa in recent years is shown in Table I, from which it is seen that the brewing of kaffir beer is an industry of considerable importance. Production has nearly trebled in the past seven years and is today nearly equal in volume to that of European ale, beer and stout which has remained steady at 20–22 million gallons per annum during this period.

**Table I**

*Production of kaffir beer in the Union of South Africa*

| Year | Volume (gal.) | Value (£) |
|---|---|---|
| 1946–47 | 7,302,000 | 667,319 |
| 1947–48 | 8,811,000 | 776,573 |
| 1948–49 | 9,798,000 | 929,714 |
| 1949–50 | 11,122,000 | 1,057,074 |
| 1950–51 | 12,292,000 | 1,181,413 |
| 1951–52 | 13,862,000 | 1,331,512 |
| 1953–54 | 20,734,000* | 2,017,274 |

* Municipal breweries only

## The malting of kaffircorn

In the Union, kaffir beer is generally made from kaffircorn malt and unmalted kaffircorn meal, but maize meal and other maize products may be substituted for part or all of the kaffircorn meal. The choice of cereal depends partly on the relative prices of kaffircorn and maize and partly on the personal preference of the brewer. In the Rhodesias other grains, such as *ropoko* or *rukwesa* (finger millet, *Eleusine coracana* Gaertn.) and *nyauti* or *munga* (bulrush millet, *Pennisetum typhoideum* Burm.) are often used instead of kaffircorn, both for the malt as well as for the meal.

The bulk of the 100,000–300,000 short tons of grain sorghums produced annually in the Union consists of kaffircorn (*Sorghum caffrorum* Beauv.); other types such as milo (*S. subglabrescens* Schweinf. & Aschers), hegari and feterita (*S. caudatum* var. *feterita* Stapf.) are cultivated only on a comparatively small scale.[8] Although the kaffircorn is grown primarily for malting and brewing, little or nothing is known of the malting qualities of the numerous varieties planted.

The production of kaffircorn malt is of the order of 90,000 short tons per annum, nearly all of which is produced by malting on concrete floors in the open. Some of the methods employed by different operators have been described by Oxford[7] and Lazar.[9] The grain is steeped in concrete tanks for 6 to 36 h. The steep water may be changed once or twice, but this is seldom done where a short steeping time is used. The grain is then spread out in beds 5–8 in. deep, which are covered with sacking and watered from time to time to keep them moist. The grain is allowed to germinate for 4–6 days. Some maltsters turn the grain daily, but in most cases it is not disturbed. No control of the temperature during germination is possible. In winter the grain is usually couched deeper and germinated longer, to compensate for the lower ambient temperature. Germination is uneven and the upper layer of the bed often germinates badly because of lack of moisture. When the germination has gone far enough, as judged by the length of the plumule, the malt is spread in thin layers in the sun to dry. Over 80% of the kaffircorn malt produced is purchased by the Bantu for home brewing, and since they judge the quality of malt by the length of the plumule, germination is generally allowed to proceed until the plumule is 1 inch or more in length. Considerable pains are taken not to break up the plumule when the malt is ground.

Several of the large municipal breweries which make their own malt have recently installed pneumatic malting equipment in order to obtain a more uniform product. The grain is first cleaned by screening to remove foreign matter and broken corns. It is then washed and steeped for 9–24 h. The steep water is generally aerated and changed at least once. The steeped grain is placed to a depth of 3–4 ft. in a concrete malting box with a false bottom through which attemperated air is passed. The beds are turned daily and are watered lightly during the first 3 or 4 days. The grain is germinated at 25–35° for 5–6 days, after which the malt is dried at 30–35° by passing warm air through the bed. With this system greater control over the conditions of malting is possible than with outdoor floor malting. Little is known, however, about the optimum conditions for malting kaffircorn. Some preliminary studies have been reported by Murray Crone[10] and White,[11] but the data are very scanty.

The diastatic power of kaffircorn malt is generally much lower than that of barley malt. Lazar[9] examined five commercial kaffircorn malts and found the diastatic power ranged from nil to 7·8° Lintner. White[11] reported values from 3 to 18° Lintner for malts made by the

Johannesburg Municipal Brewery. A survey of the diastatic power of kaffircorn malts carried out in this laboratory[12] gave the following results:

| | Number of samples | Diastatic power (° Lintner) Range | Average |
|---|---|---|---|
| Outdoor floor malting | 17 | 4–24 | 11·6 |
| Pneumatic malting | 13 | 6–21 | 17·2 |

As was to be expected, pneumatic malting gave a much more uniform product with a higher average diastatic power than outdoor floor malting. The highest diastatic power obtained so far in a malt produced in the laboratory is 55° Lintner.[12]

Because of the very low diastatic power of some kaffircorn malts, some authors have questioned the part played by the malt in the brewing of kaffir beer, and have expressed the opinion that liquefaction and saccharification of the mash are brought about by acid hydrolysis[9] or by the action of mould amylases.[4, 6] Studies in this laboratory[12, 13] have shown, however, that this is not the case and that the enzymes of the malt are sufficiently active to bring about the necessary conversion of the grain.

## The large-scale brewing of kaffir beer

The brewing of kaffir beer on a large scale is based on the traditional Bantu methods. In many municipal and mine breweries, the equipment is primitive and the brewing is carried out by men with no scientific knowledge of the process.[14] Within the past five years, however, a number of larger municipal breweries have installed stainless steel equipment and have made efforts to brew along scientific lines.

In the brewing process, a lactic acid fermentation precedes the alcoholic fermentation, whereas in European brewing this practice is absent or less frequently used than formerly. The process used some years ago at Durban municipal brewery was described by Oxford,[7] and Young[14] has described that used in Nkana in Northern Rhodesia. From these accounts it can be seen that the method of brewing may vary considerably. In most cases, however, the process consists of the following six steps: mashing, souring, boiling, conversion, straining, and alcoholic fermentation.

Mashing is usually carried out with hot water. In one brewery, for example, 540 lb. of ground kaffircorn malt and 1600 lb. of kaffircorn meal are mixed with 500–600 gal. of water at 50° (Table II). Table II also shows another recipe in which maize meal is used. There is a considerable variation in the ratio of malt to unmalted grain in the mash at different breweries. To some extent this reflects the variability of quality of the malt available at the present time.

In the older breweries, the mash is run into shallow wooden troughs and allowed to cool slowly to room temperature. Souring commences because of the proliferation of lactic acid bacteria present on the malt and grain. This flora is a very mixed one[13] and the type of organism which predominates depends largely on the temperature at which the mash is held. Under these conditions, the dominant species may differ from brew to brew, and it is difficult to produce a beer of uniform quality. In the modern breweries souring is carried out in closed stainless steel vessels held at 45–50°. This temperature favours the development of the thermophilic *Lactobacillus delbrückii* and largely suppresses the development of other lactic acid bacteria, as well as of spoilage organisms. It also greatly reduces the souring time. Whereas in the old process, souring took up to 48 h., it is now complete in 10–15 h., and under favourable conditions may even be complete in 6 hr. Inoculation with a pure culture of *Lactobacillus delbrückii* has not been employed up to the present, although it is logically the next step. Inoculation with soured mash from the previous batch is, however, practised in some breweries. As the souring progresses, the pH drops from an initial value of about 6·0 to 3·5–3·0, and the lactic acid content increases to about 1%.

When the mash has attained the desired degree of acidity, water is added and the mixture is boiled. It is then cooled to 40–60° and more malt is added (Table II). The optimum temperature for the conversion is not known and the temperatures employed vary considerably. In the older systems, cooling is usually started immediately after the malt is added. It is slow,

however, and a considerable time elapses before the mixture reaches room temperature. In the modern installations the mash is held for about 2 h. at a temperature between 40 and 60° to secure better conversion before the wort is cooled to 25–30°.

**Table II**

*Two typical recipes for brewing of kaffir beer*

| | | I | II |
|---|---|---|---|
| Mashing : | kaffircorn malt (lb.) | 540 | 360 |
| | kaffircorn meal (lb.) | 1600 | 800 |
| | maize meal (lb.) | — | 720 |
| | water (gal.) | 500–600 | 400 |
| Boiling : | water (gal.) | To 1350–1400 | |
| Conversion : | kaffircorn malt (lb.) | 720 | 360 |

In the traditional methods of brewing, alcoholic fermentation is brought about by wild yeasts introduced with the malt at the conversion stage. Recently, however, pitching with a yeast culture has been introduced in some breweries and a dried yeast preparation consisting of a strain of *Saccharomyces cerevisiae* isolated from kaffir beer is marketed for the purpose. Before or during the fermentation stage, the wort is strained to remove the husks of the grain. In most breweries fermentation is allowed to proceed for some time before straining. Straining, however, causes the beer to lose its ' head ', so the liquor is run into storage tanks where fermentation continues for a further period before the beer is sold. After fermentation for 4–8 h., the beer is ready for consumption. It is drunk in a state of active fermentation. Fermentation of the beer in the storage tanks may continue for 40 h. or longer, depending on the rate at which it is sold. The alcohol content is between 2 and 4% by weight, depending on the efficiency of the conversion stage and the duration of the fermentation. Beer sold by the municipalities may not legally contain more than 3% by weight of alcohol. The beer also contains 0·3–0·6% of lactic acid and 4–10% of solids.

The length of time the beer can be kept is limited and this is one of the biggest problems for the municipal brewers. In the older breweries the chances of microbiological contamination during processing are considerable and it not infrequently happens that a beer which is acceptable in the morning is undrinkable by the afternoon, because of the development of bacteria producing acetic acid which imparts a sharp unpleasant taste to the beer. The use of modern equipment and improved methods of brewing has done much to reduce spoilage of this kind. Even if no souring occurs, the beer goes flat if it is kept too long, due to cessation of the fermentation, and the solids, being no longer kept in suspension by the carbon dioxide, settle out. The beer is then unsaleable. The storage life of the beer can be extended considerably by keeping it under carbon dioxide pressure, and this is done at one or two municipal breweries. Refrigeration is not used because of the cost and because the beer must be warmed to room temperature before it is sold, as the Bantu do not like their beer cold. The pasteurization and bottling of beer is a development which would be welcomed by the municipal breweries, but so far no progress has been made towards this end.

The rapid industrial development of the Union of South Africa since the war with the concomitant increasing urbanization of the Bantu has changed the brewing of kaffir beer from a home art to a large-scale undertaking. This has brought the need for the use of scientific methods of brewing, as well as for raw materials of a consistently high quality. Very little is known, however, of the biochemistry of the brewing of kaffir beer and the malting of kaffircorn. In order to provide a scientific background for the development of the kaffir beer industry, this laboratory has started a study of the brewing process and of the factors influencing the quality of kaffircorn malt. The results of this work will form the subject of further communications in this series.

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# KAFFIRCORN MALTING AND BREWING STUDIES. II.*—Studies on the Microbiology of Kaffir Beer

By J. P. VAN DER WALT

The three main microbiological conversions in the production of kaffir beer have been studied, viz.: the souring of the mash by lactic acid bacteria, the alcoholic fermentation of the wort by a variety of yeasts generally present on the malt used in the conversion and, finally, the spoilage due to volatile acid formation. It was found that in breweries where no temperature control was practised during souring, a very heterogeneous lactic acid microflora developed. Eight species were recognized. In the subsequent alcoholic fermentation twenty different yeast species were isolated from beers. *Saccharomyces cerevisiae, Candida krusei* and *Kloeckera apiculata* were found to be the predominant species. The spoilage due to excessive acetic acid formation was found to be caused by the development of the common malt and beer *Acetobacter* species.

## Introduction

Kaffir beer, the traditional and national beverage of the Bantu tribes of Southern Africa, may be characterized as a fermenting, previously soured gruel, generally prepared from ground malted and unmalted sorghum.[1] A great variety of similar beverages is still prepared today by the natives throughout Africa from indigenous cereals, and kaffir beer from Southern Africa is quite comparable with 'merissa' from the Sudan, 'bouza' from Ethiopia and 'pombe' from East Africa.

The first author who paid attention to the microflora of African millet beer was Lindner[2] who described the yeast *Schizosaccharomyces pombe* isolated from a sample of pombe. Chapman & Baker[3] examined samples of 'leting' and kaffir beer which they had received from South Africa. The English authors noted the presence of large numbers of lactic and acetic acid bacteria, as well as several yeast types, in the material examined by them. They claimed to have recognized '*Saccharomyces acidi lactici*' and *Saccharomyces fragilis* which they regarded as capable of producing considerable quantities of lactic acid. However, they were intrigued that they could not demonstrate the presence of Lindner's *Schizosaccharomyces pombe* in any of the samples they had examined. Chapman & Baker made further reference to the mould flora and to the diastatic activity, in particular of the strains of *Aspergillus oryzae* isolated by them. By virtue of its high

* Part I: Preceding paper

acidity, the authors classed kaffir beer with *koumiss*, *kephir*, Belgian *lambic* and *faro*. Klein[4] examined, presumably microscopically only, samples of kaffir beer from the Witwatersrand in the Transvaal. He regarded the souring as being due to '*Bacillus acidi lactici*' and *Clostridium butyricum*, while the alcoholic fermentation was considered to be brought about by wild yeasts. During the later stages of the fermentation, he noted a rapid increase in acidity which he attributed to an increasing acetic acid flora.

Doidge[5] studied the microflora of certain South African kaffir beers in greater detail. In her studies, she clearly recognized the significant florae of the primary souring and of the subsequent alcoholic fermentation. In connexion with the souring, she isolated and described four lactic acid bacteria, two rod-shaped [one identified as *Bact. guntheri* (Leh. et Neu.), now known as *Strep. lactis*] and two Streptococci, as well as one Micrococcus capable of marked acid production. Four different yeasts were isolated, two of which she classified as belonging to the *Mycoderma* group. Doidge was, however, unable to recognize *Schizosaccharomyces pombe* in any of the beers examined by her. Even more remarkable was the fact that she failed to detect any sporogenous yeasts. The occurrence of *Mucor rouxii* in beers and on cereals used in their production attracted attention with regard to its diastatic ability. The South African author regarded *Mucor rouxii* as an important factor in the diastatic conversion during brewing. Of interest were her experiments in which she employed the isolated lactic acid bacteria strains, *Mucor rouxii* and yeasts in combination, to produce a liquid with a similar taste to that of kaffir beer from malted sorghum gruels.

Chapman[6] and Sale & Lownie[7] refer briefly to a somewhat superficial examination of the souring stage and alcoholic fermentation by Moritz, in which no attempt was made, however, to isolate any specific organism. Webb,[8] in connexion with studies on the nutritional value of kaffir beer, refers very briefly to the microflora of the brewing process and brewing materials. Special reference is again made to the amylase activity of strains of *Aspergillus flavus* and *Mucor* sp. isolated as an important factor in the saccharification of the starch.

Within recent years the brewing of kaffir beer has developed from more or less a home art to an industry of considerable economic importance. The methods of brewing, however, had until recently not advanced much beyond the primitive methods employed by the African home brewers.[1] To meet the ever-growing demand made on the municipal breweries, advances have been made in replacing the often wasteful and empirical practices by sounder and more efficient methods. With the view to possible further improvements a systematic investigation of the microbiological conversions in the production of kaffir beer has been undertaken. The purpose of this study was to determine the significant microflora of the souring stage, the alcoholic fermentation and the subsequent spoilage. Secondly, the investigation was intended to probe the possibility of improving brewing processes by the introduction of the use of pure cultures.

### *A. The lactic acid bacteria in the souring of kaffir beer mash*

#### Experimental

For the purpose of the investigation, samples of materials at the various stages in the processing of the beer were obtained from municipal breweries and also from home brewers.

Microscopical examination of a soured mash revealed a very complex microflora. Predominant numbers of non-motile rod-shaped organisms were always present besides great numbers of diplococci, while sarcinae and yeasts were generally present as well. Plectridia and clostridial forms were frequently present.

For the study of the lactic acid flora in pure culture two methods of isolation were employed:

(*a*) *Direct plating.*—Samples of mash (pH 3·5–3·7) were plated directly on:

(1) Malt agar, 1% peptone, 5% yeast autolysate, pH 6·8; incubated at 45°.
(2) Malt agar, 2% chalk; incubated at 35°.
(3) Yeast water, 2% glucose, 2% chalk, 2% agar; incubated at 30° under anaerobic conditions.
(4) Yeast autolysate, 2% glucose, 2% chalk, 2% agar; incubated at 30° under anaerobic conditions.
(5) Yeast water gelatin, 5% sucrose; incubated at 22°.

(*b*) *Plating subsequent to enrichment.*—One-ml. samples of soured mash were inoculated into 125-ml. glass-stoppered bottles containing sterile media with the following composition:

(1) Malt extract 10° Balling (sp. gr. 1·040). Incubated at 50°. After 18 h. the contents were plated on medium (a-1) and incubated at 45°.
(2) Malt extract 10° Balling; incubated at 35°. After 24 h. the contents were plated on medium (a-2) and incubated at 35°.
(3) Yeast water, 2% glucose; incubated at 30°. After 24 h. the contents were plated as described under (a-3).
(4) Yeast water, 5% sucrose; incubated at 22°. After 48 h. the contents were plated as described under (a-5).
(5) Yeast autolysate, 2% glucose; incubated at 30°. After 24 h. the contents were plated as described under (a-4).

While (a-1), (b-1) and (a-5), (b-4) are methods more specific for the isolation of *Lactobacillus delbrückii* and the slime-producing *Betacocci*, the other media were particularly useful for the isolation of mesophilic, homo- and heterofermentative lactic acid bacteria. *Lactobacillus delbrückii* was brought into pure culture by alternate plating on (a-1) and subculturing single colonies in malt extract at 50°. *Betacocci* were brought into pure culture by repeated plating on (a-3) from which acid-producing colonies were selected. For the remainder, acid-producing, catalase-negative colonies were selected and brought into pure culture by plating and incubation in anaerobic jars. Stock cultures were maintained in milk–yeast autolysate–glucose–chalk. The purity of the cultures was regularly controlled. The cultures were examined for the following properties: production of acid from glucose, catalase activity, nitrate reduction, and the Gram stain on 24-hour cultures. All non-motile, acid-producing, asporogenous, catalase-negative, Gram-positive organisms, incapable of reducing nitrate to nitrite were then examined for morphology, ability to produce gas from glucose, growth at 15°, 30°, 45° and 50°, production of acid from various polyalcohols and sugars, growth in skimmed milk, production of polysaccharides from sucrose and the optical rotation of the lactic acid produced by analysis of the zinc salt.

Fifty strains of lactic acid bacteria were studied and classified according to the monographs of Orla Jensen[9, 10] and Bergey.[11]

The souring of kaffir beer mash was studied at different temperatures. An unsterilized mash was prepared from 500 g. of kaffircorn malt meal, 500 g. of maize meal and 500 g. of kaffircorn meal mashed with 6000 ml. of water at 45° (pH of the mash 6·0–6·1). One series of glass-stoppered bottles of 125-ml. capacity was entirely filled with mash, and a second series of similar samples was prepared with the addition of a 5% v/v inoculum of an 18-hour culture of *Lactobacillus delbrückii* in sterile mash. Both series were incubated at 25°, 30°, 37°, 45° and 50° for 24 h. during which period the variation in pH was measured at four-hourly intervals. After 24 h. the contents of the bottles were examined microscopically after application of the Gram stains.

## Results

The results are recorded in Figs. 1 and 2 and in Tables I and II.

The following groups of organisms were identified in the mashes and on malts.

(1) Filamentous, Gram-positive, catalase-negative rods capable of growth at 50°, but not at 15°. No gas was produced from glucose. Acid was produced from glucose, fructose, galactose, maltose and saccharose, but not from lactose or pentose. L(+)-Lactic acid was produced from glucose. The organisms of this group were readily identified as *Lactobacillus delbrückii* (Leichmann) Beijerinck (*Thermobacterium cereale*, Orla Jensen).

(2) Gram-positive, catalase-negative rods, often occurring in chains, incapable of growth at 50°. No gas was produced from glucose. Acid was produced from glucose, galactose, sucrose, maltose, lactose, melibiose, raffinose and arabinose. The organisms grew in media containing 5% NaCl. Inactive lactic acid was produced. The organisms of this group were identified as *Lactobacillus plantarum* (Orla Jensen) Holland (*Streptobacterium plantarum* Orla Jensen).

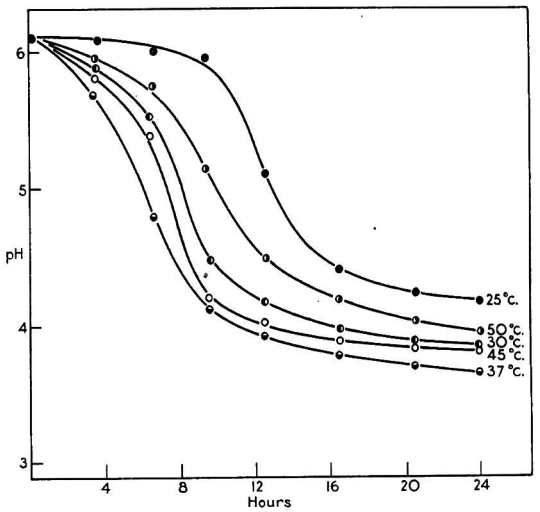


FIG. 1.—*The souring rate of unsterilized mashes souring spontaneously*

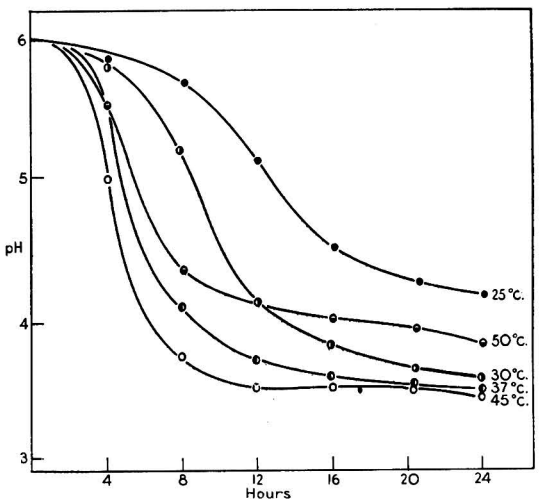


FIG. 2.—***The souring rate of unsterilized mashes seeded with a 5% v/v inoculum of*** Lactobacillus delbrückii

(3) Gram-positive, catalase-negative rods, incapable of growth at 50°. Gas was produced from glucose, but not from lactate. Acid was produced from glucose, galactose, sucrose, maltose, arabinose and xylose. Fructose was reduced to mannitol. The lactic acid produced was generally inactive. This group was identified as *Lactobacillus brevis* (Orla Jenson) Bergey *et al.* (*Betabacterium arabinosaceum* Orla Jensen).

(4) Gram-positive, catalase-negative rods, incapable of growth at 50°. Gas was produced from glucose, but not from lactate. Acid was produced from glucose, galactose, maltose, sucrose, lactose and xylose, but not from arabinose. Fructose was reduced to mannitol. Inactive

**Table I**

*The variation in microflora in mashes souring spontaneously for 24 h. at 25°, 30°, 37°, 45° and 50°*

| Temperature °c | Decrease in pH | Gram-positive rods (Mesophilic) lactic acid bacteria) | Thermophilic Gram-positive filamentous rods (*Lb. delbrückii*) | Streptococci, Pediococci and Leuconostoc spp. | Yeasts | Motile rods |
|---|---|---|---|---|---|---|
| 25 | 1·95 | + | − | +++ | + | ++ |
| 30 | 2·3 | +++ | − | +++ | ++ | + |
| 37 | 2·4 | +++ | − | +++ | ++ | − |
| 45 | 2·3 | +++ | ++ | + | − | − |
| 50 | 2·2 | − | +++ | − | − | − |

**Table II**

*The variation in microflora souring for 24 h. at 25°, 30°, 37°, 45° and 50° in mashes inoculated with 5% v/v inoculum of* Lactobacillus delbrückii

| Temperature ° c | Decrease in pH | Gram-positive rods (Mesophilic lactic acid bacteria) | Thermophilic Gram-positive filamentous rods (*Lb. delbrückii*) | Streptococci, Pediococci and Leuconostoc spp. | Yeasts | Motile rods |
|---|---|---|---|---|---|---|
| 25 | 1·9 | + | − | +++ | + | ++ |
| 30 | 2·5 | +++ | − | ++ | ++ | + |
| 37 | 2·55 | +++ | + | + | + | − |
| 45 | 2·6 | + | +++ | − | − | − |
| 50 | 2·4 | − | +++ | − | − | − |

lactic acid was generally produced. The organisms of this group could be identified as *Lactobacillus fermenti* Beijerinck (*Betabacterium longum* Orla Jensen).

(5) Gram-positive, catalase-negative diplococci with a tendency to form tetrads in neutral media. No growth occurred at 50°. No gas was produced from glucose. Acid was produced from glucose, galactose, maltose, arabinose and salicin, and very little or none from sucrose. Inactive lactic acid was produced. The strains of this group were readily identified as *Pediococcus damnosus* Clausen described by Mees.[12]

(6) Gram-positive, catalase-negative diplococci or very short rods occurring in pairs and often in short chains. No growth occurred at 50°. Gas was produced from glucose. Acid was produced from glucose, galactose, maltose, sucrose, xylose and arabinose. On media containing sucrose, slime was generally produced. The lactic acid produced was inactive. The strains could be identified as *Leuconostoc mesenteroides* (Cienkowski) van Tieghem (*Betacoccus arabinosaceus* Orla Jensen).

(7) Gram-positive, catalase-negative diplococci or very short rods, occurring in pairs or short chains, incapable of growth at 50°. Gas was produced from glucose. Acid was produced from glucose, galactose, sucrose, maltose, but not from arabinose, raffinose or glycerol. Fructose was reduced to mannitol. Generally inactive lactic acid was produced. Slime was produced on media containing sucrose. This group was readily identified as *Leuconostoc dextranicum* (Beijerinck) Hucker & Pederson (*Betacoccus dextranicus* Orla Jensen).

(8) Occasionally *Streptococcus* spp. were encountered. Since these organisms occurred only infrequently, no detailed classification was undertaken. Most strains failed to grow at 50° and produced acid from glucose, galactose, saccharose, maltose, lactose and salicin.

## Discussion

The complexity of the lactic acid flora is more or less in accordance with expectations. The lactic acid bacteria isolated, viz. *Lactobacillus delbrückii, Lb. plantarum, Lb. brevis, Lb. fermenti, Pediococcus damnosus, Leuconostoc mesenteroides* and *Leuconostoc dextranicum*, are common in all souring mashes and are introduced into the mashes by the ingredients.

Where no control of souring temperature is exercised, the predominance of any one species and the general composition of the lactic flora will vary with conditions prevailing in the brewery. The influence of temperature control is evident from Figs. 1 and 2 and Tables I and II.

The results from Fig. 1 show that with the raw materials used in the breweries and in the present experiments, the spontaneous souring of mashes proceeds most rapidly at temperatures favouring the development of the hetero- and homofermentative mesophilic lactic acid bacteria. This is in accordance with the picture obtained microscopically. The souring rate is, however, dependent on the initial meso- and thermophilic lactic acid bacteria count of the materials used prior to mashing. If this condition is altered by inoculation, a different picture is obtained, as is seen from Fig. 2 and Table II. Fig. 2 shows the variation in pH at different temperatures of the same mash which had been seeded with a 5% v/v inoculum of *Lactobacillus delbrückii*. Table II shows the change in microflora especially notable at the higher temperatures. Whereas considerable numbers of short Gram-positive rods and cocci, which may be taken as an indication of the mesophilic lactic acid bacteria, still develop at 45° in the mashes souring spontaneously, these are greatly depressed in the seeded mashes at 45°. Temperature control of unseeded mashes offer no special advantage. The combination of seeding with *Lactobacillus delbrückii* together with temperature control may, however, be applied advantageously. For the reduction of pH from approximately 6 to 4, the souring time is reduced by 50% in seeded mashes maintained at 45°. Whereas in unseeded kaffir beer mash a period of 12 hours is required for this reduction, in seeded mashes it can be effected in 6 hours. The higher souring temperature, 45°, together with the rapid increase in acidity in seeded mashes also reduces the hazard of the development of any undesired microflora in the souring mash. Another advantage of seeding at 45° is that, while the mesophilic lactic acid bacteria generally produce racemic and dextrorotatory lactic acid, *Lactobacillus delbrückii* produces the laevorotatory isomer which is preferable from the nutritional point of view, especially where large quantities of acid are consumed, as is the case with kaffir beer.

### *B. The alcoholic fermentation of kaffir beer*

#### Experimental

Representative samples of malt and beer were obtained from municipal breweries and home brewers. Twenty-five samples of beer and twelve samples of malt were examined.

Microscopical examination of a beer ready for consumption generally reveals the presence of great numbers of yeast cells, starch granules and a variety of bacteria. A normal beer has a yeast count of about $10^8$ viable cells per ml.

For the isolation of the yeasts in pure culture two methods were employed:

(*a*) Direct plating on malt agar (10° Balling unhopped malt extract pH 5·5–5·3, 2% agar). The beer was plated directly and malt samples were suspended in water. Plates were incubated at 25°. Pure cultures were obtained by subsequent plating.

(*b*) Enrichment in 15° Balling malt extract. One-ml. aliquots of beer or 1-g. samples of malt were inoculated into 30 ml. of malt extract and incubated for 24–36 hr. at 25°. These enrichments were then plated and pure cultures obtained by subsequent plating.

For the identification and classification of the isolated strains, the taxonomic study of Lodder & Kreger-van Rij[13] was employed and their standard methods adopted.

#### Results and discussion

One hundred and four strains, each a qualitative representative of each primary plate culture made, were isolated and identified.

In the traditional method of brewing, the addition of malt added for the diastatic conversion, serves also as yeast inoculum for the alcoholic fermentation. The extreme heterogeneity of a yeast flora derived from such a source is evident from Table III.

No significant qualitative differences were observed between the yeast flora of malt and that of beer. *Saccharomyces cerevisiae, Candida krusei* and *Kloeckera apiculata* were found to be dominant yeasts both in beer and malt. *Saccharomyces cerevisiae*, however, is the only organism of significance because of its numerical predominance and superior fermentative ability.

The alcoholic fermentation in kaffir beer is therefore a typical beer fermentation. It differs from the highly specialized European beer fermentation in several aspects.

**Table III**

*The yeast flora*

| Organism | Source of isolation | Number of cultures |
|---|---|---|
| *Saccharomyces cerevisiae* Hansen | Beer and malt | 26 |
| *Candida krusei* (Cast.) Berkhout | Beer and malt | 18 |
| *Kloeckera apiculata* (Reess *emend* Klöcker) Janke | Beer and malt | 11 |
| *Candida tropicalis* (Cast.) Berkhout | Beer and malt | 8 |
| *Saccharomyces carlsbergensis* Hansen | Beer | 4 |
| *Saccharomyces marxianus* Hansen | Beer and malt | 4 |
| *Hansenula anomala* (Hansen) P. et H. Sydow | Beer and malt | 4 |
| *Endomycopsis fibuliger* (Lindner) Stelling-Dekker | Beer and malt | 4 |
| *Candida robusta* Diddens et Lodder | Beer | 4 |
| *Saccharomyces steineri* Lodder et Kreger-van Rij | Beer | 3 |
| *Pichia fermentans* Lodder | Beer and malt | 3 |
| *Saccharomyces chevalieri* Gyillermond | Beer | 2 |
| *Saccharomyces willianus* Saccardo | Beer | 2 |
| *Candida macedoniensis* (Cast. et Chalmers) Berkhout | Beer | 2 |
| *Candida utilis* (Henneberg) Lodder et Kreger-van Rij | Beer | 2 |
| *Candida pelliculosa* Redaelli | Beer | 2 |
| *Torulopsis holmii* (Jörgensen) Lodder | Beer | 2 |
| *Hansenula subpelliculosa* Bedford | Beer | 1 |
| *Candida guillermondii* (Cast.) Langeron et Guerra | Beer | 1 |
| *Candida mycoderma* (Reess) Lodder et Kreger-van Rij | Beer | 1 |

In the first instance, the fermentation of the unclarified wort is allowed to proceed at atmospheric temperature (~20°) which is higher than that maintained for either the top or bottom fermentation in the European brewing industry.

Secondly, the alcoholic fermentation in the case of kaffir beer is a rapid process, complete within 8–18 h., whereas the main fermentation in the European beer industry is continued for several days and is followed by an after-fermentation.

Thirdly, whereas pitching is universally practised in the European brewing industry, this procedure has only lately been introduced in the more modern kaffir beer breweries. For this purpose, strains of *Saccharomyces cerevisiae* are employed which give a vigorous top fermentation.

Despite the great variety in the yeast flora of kaffir beer, it was remarkable that Lindner's *Schizosaccharomyces pombe* was never encountered either in beer or in enrichments from malt, although particular attention has been given to the possible occurrence of yeasts budding on a broad base. The popular belief that the alcoholic fermentation of kaffir beer would be due only to the above mentioned organisms seems to be unfounded.

Chapman & Baker[3] compared kaffir beer to *lambic* and *faro* because of its acid taste. This resemblance is, however, slight. Like *lambic*, primitively brewed kaffir beer is the product of a more or less spontaneous fermentation in that pitching is not practised. On the other hand, it differs from *lambic* in that the latter is subjected to a very long after-fermentation during which yeasts of the genus *Brettanomyces* are responsible for the typical bouquet of the beer. Several kaffir beers were examined for the presence of *Brettanomyces* according to the procedure described by Custers,[14] but without success. The reason must be sought in the slow development of these organisms which, if present on malt, are rapidly overgrown by the more robust genera.

### *C. The spoilage of kaffir beer*

On keeping of a sample of kaffir beer there is a marked change in appearance. The fermentation subsides and sedimentation of the solids takes place, leaving a clear supernatant liquid on which a pellicle eventually develops. Within 1–8 days, the pleasant sour taste due to lactic acid becomes masked by a vinegary flavour due to acetic acid formation, rendering the beer unsaleable. While lactic acid concentrations as high as 1% are tolerated, an acetic acid content of 0·5% renders a beer unacceptable. In breweries operating with primitive equipment this spoilage assumes hazardous proportions, especially in the summer months. The limited keeping quality of the beer is the major problem which confronts brewers. Chapman & Baker[3] and also Klein[4]

described the occurrence of acetic acid bacteria especially in such old beers. Klein also noted the presence of butyric acid bacteria. Excessive volatile acid production by the latter organisms is, however, unlikely due to the high acidity of the beer.

## Experimental

*Microscopical examination.*—On examination of an old beer, considerable numbers of Gram-positive and Gram-negative, motile and non-motile bacteria, and not infrequently motile clostridia were found. The pellicle which eventually formed was found to consist of long oval to elongated yeast cells, together with immense numbers of actively motile Gram-negative rods.

*Cultural.*—These organisms were readily obtained in pure culture by plating on 10% glucose, 3% chalk yeast water agar and on malt agar, incubating at 25°. The pellicle-forming yeasts were recognized by their appearance, while colonies of the acetic acid bacteria are characterized by the marked dissolution of the chalk due to acid formation.

The acetic acid bacteria obtained in pure culture were classified according to the monograph of Frateur,[15] while the pellicle-forming yeasts were identified according to Lodder & Kreger-van Rij.[13]

The clostridial forms present were isolated by inoculation of 1-ml. samples of beer into 125-ml. stoppered bottles containing 2% glucose, 1% chalk yeast water, and incubation at 35° for 4–5 days. After spore formation, pasteurized samples of the contents were plated on 2% glucose, 0·5% chalk yeast water agar, and the plates incubated anaerobically gave pure cultures of acid-forming Clostridia.

## Results and discussion

The *Acetobacter* cultures were found to correspond to *Acetobacter rancens*, *Acetobacter ascendens* and occasionally *Acetobacter suboxydans*. The pellicle-forming yeasts could invariably be identified with *Candida krusei* and *Candida mycoderma*. Less frequently strains of *Pichia membranefaciens* and *Hanseula anomala* were encountered. The cultures of acid-producing Clostridia corresponded to *Clostridium butyricum* Prazmowski.

Both the pellicle-forming yeasts and the common beer *Acetobacter* species possess an oxidative dissimilation, developing chiefly on the alcohol and lactic acid present. In the older breweries these organisms are generally introduced into the more or less sterile boiled porridges by the addition of the malt for diastatic conversion. Concomitantly lactic acid bacteria are again introduced which are, however, capable of a limited activity. Excessive volatile acid formation is due to the development of *Acetobacter* species.

The presence of mould growth on the surfaces of cooling or souring porridges is no uncommon occurrence in breweries still employing open wooden vessels. In extreme cases a felted mould growth spreads over the surface. This condition is greatly influenced by atmospheric conditions. With low atmospheric temperatures, souring is retarded and prolonged exposure of the mash facilitates the infection. In extreme cases the porridge in such infected vessels shows a tendency to spontaneous liquefaction.

The occurrence of amylolytic micro-organisms on cereal products is common. Chapman & Baker,[3] Doidge[5] and also Webb[8] paid considerable attention to the diastatic activity of the mould flora on cereals employed in the preparation of kaffir beer. Webb concluded from his investigations that the low diastatic activities of germinated maize and sorghum in themselves were too low to account for the sugar production to attain the alcohol contents found in practice. In sterilized maize mashes inoculated with pure cultures of *Aspergillus flavus* isolated from brewing materials, 5% sugar (as glucose) was obtained in 19 hours.

In the present investigation, a cursory survey showed the most common moulds present on kaffircorn malt to be *Mucor* spp., *Rhizopus* spp. and *Aspergillus* spp., while only occasionally *Penicillia* and *Oospora* spp. were encountered.

Cultures of *Endomycopsis fibuliger* were frequently isolated from malt samples. Studies by Wickerham *et al.*[16] have shown that this yeast is a potent diastatic agent under favourable conditions.

A certain limited degree of saccharification due to mould amylase seems feasible. This activity has, however, been overestimated by previous investigators who have based their conclusions on pure-culture experiments in sterile mashes—conditions not representing true practice. No mould preparations are used in the manufacture of kaffir beer, as is the case with oriental beverages such as *saké*. The diastatic activity of commercial kaffircorn malts is known to be low, generally due to faulty malting procedures and the use of unsuitable varieties of kaffircorn. Despite the low activity of the malts, conversion is brought about chiefly by cereal amylase, since the mould counts of satisfactory malts and those of malts of very low diastatic activity were not found to differ appreciably.

## Conclusions

The brewing of kaffir beer involves two microbiological conversions.

Examination of the microflora involved in the souring of the mash showed that it conformed to the typical lactic acid flora encountered in any souring cereal mash. Unsterilized mashes, as prepared in the breweries, may be soured most efficiently by the combination of seeding with cultures of *Lactobacillus delbrückii* and the maintaining of souring temperature at 45°.

The alcoholic fermentation under primitive brewing conditions is brought about by the natural yeast flora present on kaffircorn malt. Despite the heterogeneity, *Saccharomyces cerevisiae* is the predominant species. Pitching with selected strains of *Saccharomyces cerevisiae* producing vigorous top fermentation has already become a common practice in this industry.

The spoilage and limited keeping value of kaffir beer due to the formation of volatile acid is caused by the development of the typical beer and malt *Acetobacter* species.

Despite the occurrence of amylolytic micro-organisms on kaffircorn malt, the diastatic conversion is brought about by cereal amylases.

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# THE LOSS OF CAROTENE FROM DRIED GREEN CROP DURING STORAGE.—The Gradient of Loss through a Stack

By V. H. BOOTH*

Considerably more carotene was lost from the middle of a commercial type stack of dry lucerne meal than from the edges. The percentage loss, small near the outermost edges of paper sacks which were exposed to air, rose sharply in the first few cm. and then more gradually to the centre of the stack. Part of the increased loss in the centre was due to the higher temperature there. The major part of the difference in the rates of loss was due to the protective effect of moisture taken up from the air at the outer surfaces of the stack. These conclusions were verified by isolating sections of stacks of dry meal in tubes 2½ metres long closed at one end and so stored as to eliminate temperature gradients. Loss of carotene was high within a tube except near the exposed end where moisture had been taken up.

## Introduction

Dried green crop meals are produced in season, stored, and used all the year round. They are used, for instance, in pig and poultry mashes to supply vitamin A in the form of carotene. The meal is normally stored in paper sacks built into stacks inside darkened warehouses, the stacks usually containing between 10 and 100 tons. During a typical storage period of six months, a dried green crop meal loses by oxidation a quarter to a half of its carotene. Many factors—light, temperature, moisture, antioxidants and other additives, type of packing, degree of compression—which affect or control the rate of loss of carotene have been studied with small isolated specimens of meal. A review of these factors and a list of references are given in a monograph.[1] The rate of loss of carotene from different parts of a whole stack does not appear to have been studied, partly no doubt because the scale of an experiment brings difficulties both of cost and of sampling. The results of such an investigation are described in the present paper, in which it is shown that the loss of carotene was greater in the middle of a stack than at the outer edges, and that the factors concerned in this difference are partly temperature but chiefly moisture content.

## Experimental

### *Methods*

Even in a well-run drying plant, the meal as produced varies continually in quality. Sometimes a difference in colour is easily visible between different sackfuls from the same batch. Therefore in these experiments enough meal to fill all those sacks which were later analysed was mixed before bagging. A specimen of the mixed meal was analysed for carotene content by extracting with hot light petroleum according to Duodecim Viri.[2] The pigment solution so obtained was purified chromatographically on a mixture of aluminium oxide and anhydrous sodium sulphate,[3, 4, 5] and the carotene in light petroleum solution determined in a Unicam Glass Photoelectric Spectrophotometer (Model SP600) at 450 mμ. After storage of meal, the contents of a sack or zone were mixed and similarly analysed. Determinations on every specimen of meal were done in triplicate. This method is chosen for its simplicity, although in our experience a small amount of carotene remains unextracted and another small amount is destroyed. The standard error of the mean of triplicates, based on results with 200 triplicates having mean values between 140 and 280 p.p.m., was 2·0 p.p.m. This low scatter value suggests that the destruction is nearly constant and, as all the work was comparative, the small loss has been neglected.

Moisture determinations were made with a Marconi moisture meter. This is an electronic instrument designed primarily for grain. It was calibrated by taking readings on meals whose moisture contents were determined by a heating method. The precision is a little lower than with the usual weighing and heating methods and the results may be up to 1½% units in error. The method is rapid and therefore many observations were made including exploratory ones.

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The instrument is portable and observations were made in the warehouse immediately each sack was opened. Without such a method the main experiment reported herein would have been very much more complicated.

(1) *Experiments with stacked sacks of meal*

*The first stack experiment*

In a preliminary experiment, a stack of 10 tons of lucerne meal contained in 400 sacks included 14 experimental sacks in the middle and 14 at the outside corners. After 27 weeks the contents of each experimental sack were separately mixed and analysed in triplicate. From the sacks in the centre of the stack, 55% (± 0·69*) of the carotene was lost, against only 40·6% (± 0·99) from the corner sacks. The difference was statistically significant (P = <0·001). Because it is well known that the rate of destruction of carotene increases with temperature, the meal had been cooled before bagging and stacking, in June. At the time of breaking the stack, in December, the temperature of the air both within and without the stack was 12·5°. Therefore the differential destruction of carotene is unlikely to have been entirely due to a temperature gradient.

*Control of moisture*

Dried green crop meal as produced has a moisture content in the neighbourhood of 5%. In equilibrium with air at 70% r.h. the moisture content is about 12%. The moisture, although quickly taken up at the exposed surface of meal, penetrates only slower into deeper layers.

Bielefeldt,[6] and others since, stored dried green crop meal in a series of sealed vessels each having a different moisture level. Carotene loss was least in presence of the greatest amount of moisture, apparently because at the higher moisture levels micro-organisms were more active in using up the limited oxygen. In commercial practice, green crop meals are stored in paper sacks and therefore access of oxygen is not a limiting factor. Of more practical interest, therefore, are the experiments of Bailey, Atkins & Bickoff[7] who stored specimens of lucerne meals at different moisture levels in a series of humidistats containing excess air and maintained at 40°. They found, as earlier work by Halverson & Hart,[8] Silker, Schrenk & King[9] and Mitchell, Schrenk & King[10] had suggested, that carotene was best preserved in meal having a moisture content of about 6–7%. No satisfactory hypothesis appears to have been proposed to explain the mechanism of the action of moisture in presence of unlimited air.

A simpler way of carrying out this experiment at different moisture levels is to add various amounts of water to specimens of dry meal and to store them in sealed bags made of polythene. Polythene is sufficiently permeable to oxygen for this to be no limiting factor,[1] yet sufficiently impermeable to water vapour for the present purpose. When a series of such specimens was stored for 16 weeks at 26°, similar results were found to those of Bailey *et al.* but the optimum moisture was a little higher, namely at about 8%. This small difference must have been due to some difference in storage conditions such as lower temperature. In the preliminary stack experiment, moisture determinations were not made, but as dried green crop meal is often produced with lower moisture contents than this optimum value and as meal is known to take up moisture from the air, it was considered probable that the carotene at the outside of the stack received protection through greater uptake of moisture there.

*The second stack experiment*

The stack experiment was therefore repeated. Temperatures were recorded throughout the storage period and moisture contents were determined at the end of the experiment. Some 14 tons of lucerne meal in paper sacks each containing ½ cwt. (25 kg.) were cooled. The contents of 24 sacks were mixed, part was refilled into 13 paper sacks and part into nine bags of polythene having a capacity of about 4 kg. each. The carotene content was 270 p.p.m. and the moisture content was about 5%. Extra water was sprayed on to another portion of the mixed meal to bring the moisture content to about 10%, and this was filled into two paper sacks and nine polythene bags. The 18 polythene bags were closed by heat sealing and placed inside paper bags.

* Standard error of the mean

An almost cubical stack was built with the 14 tons of meal in sacks within a darkened warehouse. The 13 experimental sacks and 18 smaller bags were distributed in the two middle courses in a manner which is partially shown in plan in Fig. 1. Each was replicated but replications are omitted from the figure to avoid confusion. The two paper sacks of damped meal were placed on top of the stack. The stack was built on 30th October and remained undisturbed until 24th March.

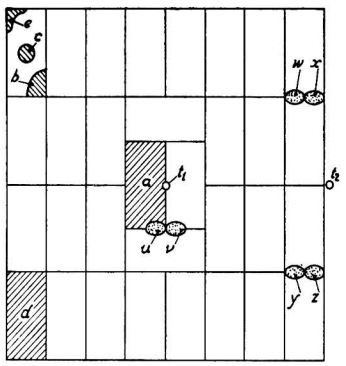


FIG. 1.—*Formalized and simplified plan of middle courses of sacks in stack*

Thermometers were placed at $t_1$ and $t_2$. For explanation of other symbols see Table I. The smaller bags distorted the courses slightly: the distortions were unimportant and are not shown. All positions were replicated.

When the stack was dismantled, the contents of each sack, bag or section of a sack were mixed. The sections *b*, *c* and *e* were made by cutting vertically through the paper sack plus contents with a knife. Moisture determinations were made immediately and carotene determinations were made from 3 to 5 days later. The results are shown in Table I. The principal results are marked with asterisks: the other results represent intermediate positions in the stack. All the 18 polythene bags were used for moisture observations, but only 13 were used for carotene analysis. Agreement between replicates was good as shown by the pooled standard deviation of one replicate, namely 3·1 in % loss units. Because this agreement was good and because the differences between types and positions were clear cut, it was considered unnecessary to analyse the other five replicates for carotene content.

## Table I

*Moisture contents and carotene losses in different parts of a stack of meal stored five months in winter*

| Position in stack† and packing material | Replications‡ | Moisture, % | Carotene, % lost |
|---|---|---|---|
| **a* whole contents of middle paper sack | 4 | 5 | 42·6 |
| *b* inner corner of outside paper sack | 3 | 6 | 20·2 |
| *c* middle of outside paper sack | | 6 | |
| *d* whole contents of outside paper sack | 3 | 8 | 16·4 |
| **e* outer corner of outside paper sack | 3 | 13 | 8·3 |
| *f* top of stack, ' wet ' meal in paper sack | 1 | 12 | 10·1 |
| **u* middle polythene bag, ' dry ' meal | 2 | 5 | 37·6 |
| *w* nearly outside polythene bag, ' dry ' meal | 2 | 4 | 30·5 |
| **z* outside polythene bag, ' dry ' meal | 2 | 5 | 26·0 |
| **v* middle polythene bag, ' wet ' meal | 2 | 8 | 13·2 |
| *y* nearly outside polythene bag, ' wet ' meal | 2 | 9 | 8·5 |
| **x* outside polythene bag, ' wet ' meal | 3 | 8 | 7·5 |

* At first reading only the six starred results should be studied. They represent extreme positions
† For position see Fig. 1. Sacks *f* are not shown; they lie flat on top of the stack
‡ Number of separate sacks, bags or sections used for carotene analysis

### *The part played by temperature gradient*

One remote-reading thermometer was placed in the centre of the stack and another outside but close to the stack. Temperatures were observed at noon. Because the daily temperatures outside the stack fluctuated widely, the averages for groups of 12 days are shown in Table II.

## Table II

*Temperatures within and without the stack: averages for groups of 12 days throughout storage*

| Position | Temperature, °F | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Inside, $t_1$ | 66·2 | 66·0 | 64·2 | 62·6 | 60·1 | 58·7 | 56·0 | 53·8 | 52·2 | 50·7 | 49·4 | 48·0 |
| Outside, $t_2$ | 55·1 | 49·9 | 49·6 | 49·0 | 50·7 | 47·0 | 35·8 | 46·9 | 42·8 | 34·3 | 36·8 | 42·0 |
| Difference | 11·1 | 16·1 | 14·6 | 13·6 | 9·4 | 11·7 | 20·2 | 6·9 | 9·4 | 16·4 | 12·6 | 6·0 |
| Mean differences | 13·9 | | | | 12·0 | | | | 11·1 | | | |

The temperature within the stack fell steadily. The differences between group averages for the inside and outside temperatures were almost steady and the overall average difference was 12·3° F (6·8° C). The average distance from the thermometer in the middle of the stack to the outside was a little under 2 metres. In the first stack experiment the meal was cooled before bagging, and in the second, after bagging. The latter is not so effective and this doubtless accounts for the temperature inside the sacks being above that of the air.

No exact study of the kinetics of the loss of carotene appears to have been published although Kohler, Beier & Bolze[11] refer to unpublished work. The impression gained from the literature (for references see Booth[1]) is that the loss is approximately logarithmic except at the beginning and towards the end. The temperature coefficient is in the neighbourhood of 2 (Kohler *et al.*[11]). The extreme temperature difference across the stack was 6·8°. Positions *a* and *e* are not extreme but they are almost so. The 42·6% loss of carotene in the centre of the stack would therefore probably be reduced to about 25% at the outer edge when corrected for temperature alone. In fact the observed loss in position *e* was only 8·3%.

*The part played by moisture*

The moisture content was high in meal at the outwardly-turned edge of each paper sack and fell rapidly towards its inner edge (position *b*): the outside layer was hard while the meal 5 or 10 cm. inside was powdery. The loss of carotene was least where moisture content was highest. Both moisture contents and carotene values of the inside portions of outside sacks (position *b*) were between the extreme values for extreme positions. The high moisture content at position *e* was above the optimum for retention of carotene but the last few per cent units of moisture may only have been taken up slowly. In other words, the moisture content at the outside edges of the stack may have been near the optimum during much of the storage period. On the other hand the moisture contents in the body of the stack were below the optimum for retention of carotene.

By using bags of polythene it is possible to store ' dry ' meal at the outside of a stack and ' wet ' meal at the middle, thereby enabling the effects of moisture and of position in the stack to be disentangled. This was done and the results, which are included in Table I, give support to those obtained with paper sacks as the following comparisons show.

1. Dry meal in polythene in the middle (position *u*) lost about as much carotene as that in paper sacks (position *a*).

2. The carotene in ' wet ' meal at the outside in paper (position *e*) and in polythene (position *x*) was well preserved: the two values for the percentage loss, 8·3 and 7·5%, are of the same order as each other and as that, 10·1%, in ' wet ' meal in paper sacks on top of the stack (position *f*).

3. ' Wet ' meal in the centre and outside positions of the stack (positions *v* and *x*) lost 13·2 and 7·5% of carotene (ratio 1·76). ' Dry ' meal in the centre and outside positions (*u* and *z*) lost 37·6 and 26% carotene, respectively (ratio 1·45). The principal known difference between the members of the pairs is one of temperature. The average of the two ratios, namely 1·65, gives a rough estimate of the effect of the 6·8° C difference in temperature. The losses in nearly outside positions *w* and *y* were a little higher than the outside positions *z* and *x* and this difference is consistent with a temperature effect.

4. ' Wet ' and ' dry ' meal in the centre positions of the stack (*v* and *u*) lost 13·2 and 37·6% carotene, respectively (ratio 2·85). ' Wet ' and ' dry ' meal at the outside positions of the stack (*x* and *z*) lost 7·5 and 26% carotene, respectively (ratio 3·45). The average of these ratios is 3·15. This last value represents a rough estimate of the protective effect of moisture.

*Discussion of distribution of losses in the stack*

If we combine the temperature effect (ratio 1·65) and the moisture effect (ratio 3·15), both derived above from results on meal with controlled moisture content within polythene, we obtain $1{\cdot}65 \times 3{\cdot}15 = 5{\cdot}2$. If we divide the loss in the middle of the stack by this coefficient we obtain the value $42{\cdot}6/5{\cdot}2 = 8{\cdot}2\%$, which compares with 8·3% observed for the loss of carotene at position *e*. [The assumption is here implied that temperature and moisture are independent, an

assumption which may not be strictly correct. Further, the direct comparison of percentage losses for logarithmic rates only gives rough estimates. No more is claimed.]

The carotene was well preserved only in the outer edges of outside sacks. It will be shown below that moisture penetrates only a few inches, beyond which the loss of carotene rises. This is shown indirectly by the observation that the loss from a whole outside sack, namely 16·4% (position *d*), is nearer to that for the dry inner corner of a sack (position *b*) than for the wet outer corner. The variation between the replicate results for samples from position *d* was much greater than that between any other set, and this is understandable since the contents of each sack were far from homogeneous as regards moisture content. In the first stack experiment the extreme positions corresponded only with positions *a* and *d* and with the temperature difference probably small. With this in mind, the two experiments are seen to be in qualitative agreement.

*Application to commercial practice*

It is tentatively concluded from the results obtained with sacks *e* and *f* that if the whole of this stack could have been stored at warehouse temperature and humidity, the loss of carotene would have been only 10%, which is commercially negligible; or that by merely raising the moisture content of cooled meal the loss even at the worst position in the stack—shown by results at position *v*—would only have been 13·2% which is commercially acceptable. This storage period began in autumn and continued through the winter. Had the experiment been done in the summer all the losses shown in Table I would have been greater, but there is no reason to suppose that the conclusions would have been different.

(2) *The loss gradient in long tubes*

As experiments with whole stacks are elaborate and inconvenient, and it is difficult to allow for temperature effects, the problem has been further studied in another way. A long tube full of meal can be considered as an isolated cylindrical section of a stack. Meal at the closed end corresponds with that in the centre of the stack and meal at the mouth of the tube corresponds with that at the edge of the stack. The walls can be of glass or metal so that oxygen and moisture can only reach the closed end by migration through a length of meal as in a stack. Glass is preferred for smoothness, observation of evenness of packing, and ease of cutting. Light can be excluded and temperature differences can be avoided.

Oversize test-tubes were made by sealing one end of glass tubes 2½ metres long by 4 cm. diameter. Two tubes were filled with dried grass meal having a moisture content of 3% and a carotene value of 260 p.p.m., and the open ends were lightly stoppered with cotton wool. The tubes were wrapped in brown paper and stored horizontally out-of-doors under cover with open and closed ends opposed.

The tubes were turned occasionally during storage to obviate the effects of possible temperature gradients. After storage for 28 weeks (early July to January), the tubes were divided with a glass cutter into sections or zones. The contents of each zone were mixed, and moisture and carotene contents were determined in triplicate. Zone *a* contained the meal in the 8 cm. next the mouth of the tube, zone *b* that in the next 16 cm. and zones *c*, *d*, *e* covered about 16 cm. at equally spaced intervals down to the closed end. The mean values of the results with the two tubes are shown in Table III. Meal near the mouths of the tubes had taken up moisture from

**Table III**

*Relationship between penetration of moisture and loss of carotene in meal stored in tubes open at one end*

| Zone | Distance* cm. | Moisture % | Carotene lost, % |
|---|---|---|---|
| *a* | 4 | 10·5 | 37·4 |
| *b* | 16 | 4·9 | 42·8 |
| *c* | 85 | 3 | 55·1 |
| *d* | 155 | 3 | 52·8 |
| *e* | 230 | 3 | 54·7 |

* From open end of tube to middle of zone

the air but the moisture had not penetrated far. Carotene loss was least where moisture content was greatest and was greatest where the meal remained dry.

Other tubes were filled with meal with higher moisture content (8%) from a different source and stored indoors for 27 weeks at an average temperature of 19°. After dissection into four zones, the contents were analysed as described above. The % losses in carotene, going from open to closed ends, were (moisture content % in parentheses): (7·5) 42·1%; (8·0) 42·9%; (8·0) 43·5%; (8·1) 43·9%. The losses cannot be directly compared with those in Table III because meal and storage periods were different and the average temperature was higher, but it is evident that, while moisture contents remained approximately constant, the loss of carotene was the same throughout the length of a tube. The moisture content of the meal remained steady even near the open ends of the tubes because meal with about 8 or 10% moisture content is in equilibrium with air in a heated room.

## Conclusions

Dry meal at the outer edges of stacks and at the open ends of tubes, each exposed to air, took up moisture and lost relatively little carotene during storage, whereas the inner contents of both stacks and tubes remained dry and lost more carotene. Thus the results with long tubes qualitatively confirmed those with stacks. There were quantitative differences which may have been due to different storage conditions. In particular, moisture may have reached or passed beyond the optimum concentration at different rates in sacks separated from air by paper and in tubes in which it was separated from air by a plug of cotton wool. Several other experiments showed that the greatest distance that measurable amounts of moisture penetrated through meal during six months' storage was 22 cm. or half a sack width. The distance to which enough moisture penetrates to have a practical protective effect on carotene is only about a third of this. No difference was observed between meals made from dried grass or dried lucerne.

Dried green crop meal is frequently produced with moisture contents lower than the optimum. The managers of grass-drying plants may not realize how much lower because the moisture determinations are made in a laboratory after a lapse of time during which the small cold samples take up moisture from the air. The conclusion drawn from this work is that carotene would be much better preserved in storage if the green crop were not dried to so low a moisture content. This raises problems of moisture control and milling which are too complex to be discussed here. Suggestions for overcoming them are made elsewhere.[1]

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# THE PHOTOCHEMICAL OXIDATION OF ASCORBIC ACID IN SOLUTIONS CONTAINING OXALIC ACID. II.*—Mechanism of the Reaction

By L. H. LAMPITT, L. C. BAKER and E. WITTENBERG

Ascorbic acid is not oxidized (thermally) by ferric salts in oxalic acid solution. When ferric salts in oxalic acid solution are irradiated in an atmosphere of nitrogen, ferrous ions are produced; in an atmosphere of air, ferrous ions are produced when the ratio of iron to oxalate exceeds a certain value. It is suggested that the photochemical oxidation of ascorbic acid is due to the activity of the free radicals formed in solution on autoxidation of the ferrous ions produced by the photochemical reduction of ferri-oxalate.

## Introduction

In Part I[1] of this series it was demonstrated that light from a mercury vapour lamp promoted the oxidation of ascorbic acid in oxalic acid solution at pH 1·7 when a trace of iron salt was present and when air was passed through the solution. Under the conditions of the experiment the oxidation proceeded at a constant rate, but the actual rate increased as the iron concentration increased from 0·1 to 10·0 $\mu$g. of iron/ml. The concentration of iron therefore appeared to be a limiting factor in the rate of oxidation and it was considered probable that the constant rate of oxidation of ascorbic acid depended upon the repeated reduction and oxidation of the iron present. It has been shown by Strohecker & Sierp[2] and also by Sabalitschka[3] that ascorbic acid in dilute sulphuric acid solution is oxidized by ferric salts. However, it will be shown that in oxalic acid solution this reaction does not take place. It is suggested that the first step in the photochemical oxidation of ascorbic acid is the photochemical reduction of ferri-oxalate to ferrous salt. The sensitivity of oxalates to light is well known. According to Murray,[4] the blue-printing process, which has been known for a long time, is essentially a photochemical reduction of ferric ammonium oxalate to the ferrous salt which reacts with a soluble ferricyanide to give the blue colour; this photochemical reduction, it may be noticed, takes place in the presence of air.

The photochemical decomposition of solutions of potassium ferri-oxalate was studied by Allmand & Webb[5] and by Allmand & Young;[6] these authors found that ferrous oxalate and carbon dioxide were produced.

$$2K_3Fe(C_2O_4)_3 \rightarrow 3K_2C_2O_4 + 2FeC_2O_4 + 2CO_2$$

Potassium oxalate (0·09M) was added to the solution of potassium ferri-oxalate (0·03M) for irradiation in order to prevent the precipitation of ferrous oxalate which is not very soluble in water. When carbon dioxide was passed through the neutral solution during irradiation, ferric ions were lost (determined by titration with titanous chloride); when oxygen replaced carbon dioxide no loss of ferric ions occurred although the production of carbon dioxide could be detected. Livingston,[7] who used a similar system except that the pH of the solution was about 4 (0·02M-sulphuric acid, 0·0193M-potassium ferri-oxalate and 0·0962M-sodium oxalate), obtained similar results, namely the production of ferrous ions in the absence of oxygen but not in its presence; in the presence of oxygen, however, it was again shown that oxalate was destroyed and carbon dioxide formed. Parker[8, 9] also studied this reaction, but, using solutions which were definitely acid, he determined the ferrous ions formed with 1 : 10-phenanthroline. He found that with a solution of 0·006M-potassium ferri-oxalate in 0·1N-sulphuric acid it made no difference whether oxygen or oxygen-free nitrogen was passed through the solution during irradiation, for in each case ferrous ions were produced.

There is therefore evidence that, in the absence of oxygen and under various pH conditions, ferric ions and oxalate are lost and carbon dioxide and ferrous ions are formed, whereas, in the presence of oxygen, oxalate is lost and carbon dioxide formed, but the loss of ferric ions and the

* Part I: *J. Sci. Fd Agric.*, 1955, **6**, 682

appearance of ferrous ions depends upon the pH and upon the oxalate concentration. The behaviour of ferric ions and of oxalate under the conditions of experimentation used in Part I has therefore been investigated.

## Experimental

The standard iron solution (0·25% oxalic acid, 0·02% ascorbic acid and ferric ammonium sulphate to give 1 $\mu$g. of iron/ml.) frequently used in the experiments described in Part I of this series differs from the solutions used by the workers mentioned in the study of the photochemical decomposition of ferri-oxalate in certain important respects. The standard iron solution contains ascorbic acid and has a low ratio of iron to oxalate; furthermore the pH is notably lower, with the exception of certain of the solutions used by Parker.

The conditions of experimentation were those described in Part I.

(1) *The photochemical reduction of ferric ions in an atmosphere of nitrogen*

Nitrogen was passed through aliquot portions of a solution at pH 1·7 containing 0·25% oxalic acid, 0·02% ascorbic acid and 25 mg. ferric ammonium sulphate (+ 24 $H_2O$) per 250 ml. (2·9 mg. of iron per 250 ml.); after keeping for 1½ hours in the dark the mercury vapour lamp was switched on. After 1½ hours' irradiation, the nitrogen was replaced by air. Samples of the solutions were titrated at intervals with a solution of 2 : 6-dichlorophenol-indophenol; the results are given in Table I.

**Table I**

*Reduction of ferric ions in an atmosphere of nitrogen*

| Conditions | Total time in hours | Apparent ascorbic acid (mg. per 250 ml.) | | | | |
|---|---|---|---|---|---|---|
| | | Expt. I | Expt. II | Expt. III | Expt. IV | Average |
| Dark–nitrogen | 0 | 49·7 | 50·3 | 50·3 | 50·0 | 50·2 |
| | ½ | 50·3 | 50·3 | 50·4 | 50·7 | 50·4 |
| | 1 | 50·7 | 50·4 | 50·7 | 50·4 | 50·5 |
| | 1½ | 50·7 | 50·3 | 50·3 | 50·1 | 50·3 |
| Light–nitrogen | 2 | 56·0 | 55·4 | 54·7 | 55·4 | 55·4 |
| | 2½ | 56·7 | 55·4 | 55·7 | 56·0 | 56·0 |
| | 3 | 56·7 | 56·0 | 56·0 | 56·0 | 56·2 |
| Light–air | 3½ | 42·2 | 42·5 | 43·0 | 43·5 | 42·8 |
| | 4 | 27·7 | 28·4 | 27·0 | 32·5 | 29·2 |

The dye titration remained unchanged in the dark and increased to a more or less constant figure when the light was switched on, indicating an apparent increase in the amount of ascorbic acid present; when air replaced nitrogen a rapid loss of apparent ascorbic acid occurred. The increase in the content of apparent ascorbic acid is attributed to the photochemical reduction of ferric to ferrous ions. Lorenz & Arnold[10] used ferrous compounds for standardizing solutions of 2 : 6-dichlorophenol-indophenol; they found that in the presence of most acids (i.e., citric, acetic, phosphoric, sulphuric, nitric, hydrochloric and trichloroacetic) little, if any, reduction of the dye occurred, and that only slowly, but that in the presence of oxalic acid (and also metaphosphoric acid) reduction of the dye was rapid. Furthermore 1 mole of dye (290 g.) was reduced by 2 atoms of iron (111·7 g.) and 1 mole of dye is also reduced by 1 mole of ascorbic acid (176 g.). In terms of reducing the dye therefore:

176 g. of ascorbic acid ≡ 111·7 g. of ferrous iron

or 1 g. of ascorbic acid ≡ 0·634 g. of ferrous iron.

The increase in the amount of apparent ascorbic acid observed (5·7–7·0 mg.) is therefore equivalent to the production of 3·6–4·4 mg. of ferrous iron compared with the 2·9 mg. of ferric iron actually used.

If this interpretation of the results is correct, then it follows that no oxidation of ascorbic acid by ferric ions occurred initially in the oxalic acid solution. Had the ferric ions oxidized an equivalent amount of ascorbic acid initially in the dark, no change in dye titration would have occurred, as was in fact observed, but, in that event, on irradiation no increase in titration would be expected because the iron had already been reduced to the ferrous state.

(2) *The influence of oxalic acid on the reaction between ferric ions and ascorbic acid*

In this experiment the behaviour of two solutions was compared, the important difference between them being that in one solution the ferric salt and ascorbic acid were dissolved and the initial titration was carried out in the absence of oxalic acid, whereas in the other, oxalic acid was present in solution when the ferric salt and ascorbic acid were dissolved.

Solution I was prepared by dissolving 50 mg. of ascorbic acid and 43·3 mg. of ferric ammonium sulphate (5·0 mg. of ferric iron) in 250 ml. of dilute sulphuric acid adjusted to pH 1·7. An aliquot portion of the solution was titrated with the dye and to the remainder sufficient oxalic acid was added to give a concentration of 0·25%. Aliquots of the solution containing oxalate were then irradiated in an atmosphere of nitrogen.

In preparing solution II, 50 mg. of ascorbic acid were dissolved in 250 ml. of 0·25% oxalic acid solution at pH 1·7 and 43·3 mg. ferric ammonium sulphate subsequently added. An aliquot portion of this solution was titrated and a stream of nitrogen was passed through the remainder while it was being irradiated. The results obtained when aliquot portions of the irradiated solutions were titrated at intervals are given in Table II.

The interpretation of these results is that in Solution I, in which oxalic acid was initially absent, the ferric salt was reduced to ferrous salt by the ascorbic acid, and the ferrous salt formed, in the absence of oxalic acid, did not reduce the dye—therefore 40 mg. of ascorbic acid were found compared with the 50 mg. originally present. When the oxalic acid was added, the dye titre rose by an amount equivalent to 8 mg. of ascorbic acid or 5·07 mg. of ferrous iron, compared with 5·0 mg. of ferric iron originally used. When this solution was irradiated, no further increase in titre was observed because the iron was already in the ferrous state. In Solution II in which oxalic acid was present initially, the ferric salt was not reduced to ferrous salt because, on subsequent irradiation in nitrogen, the titre rose by an amount equivalent to 8·5–9·7 mg. of ascorbic acid or 5·4–6·17 mg. of ferrous iron. These deductions were confirmed by testing the solutions for ferric ion with potassium thiocyanate* and for ferrous ions with *o*-phenanthroline.

**Table II**

*Effect of oxalate on the reaction between ascorbic acid and ferric salt*

| Time of irradiation, hours | Apparent ascorbic acid, mg. per 250 ml. of solution | | | |
|---|---|---|---|---|
| | Solution I — Oxalic acid absent initially | | Solution II — Oxalic acid present initially | |
| | Expt. 1 | Expt. 2 | Expt. 3 | Expt. 4 |
| 0 | 40·0 | 40·0 | 50·0 | 50·0 |
| | After addition of oxalic acid | | | |
| 0 | 48·0 | 48·0 | | |
| | Irradiation in an atmosphere of nitrogen | | | |
| ½ | 48·0 | 48·0 | 59·7 | 59·0 |
| 1¼ | 48·5 | 48·7 | 58·5 | 58·5 |

| Colour reactions | KCNS | *o*-phenanthroline | KCNS | *o*-phenanthroline | KCNS | *o*-phenanthroline | KCNS | *o*-phenanthroline |
|---|---|---|---|---|---|---|---|---|
| Dark | − | + | − | + | + | trace | + | trace |
| Light | − | + | − | + | − | + | − | + |

It may be concluded from this and the previous experiment that, in an atmosphere of nitrogen, ferric salt oxidizes ascorbic acid and is reduced to ferrous salt in sulphuric acid solution but not in oxalic acid solution; in oxalic acid solution ferrous salt is formed on irradiation. When air was passed through the solution containing photochemically produced ferrous ions, there was a rapid fall of dye titre (see Table I) but whether this fall was due to loss of ascorbic acid only or ascorbic acid and ferrous ions remains to be elucidated.

* It was necessary to acidify the solution strongly to obtain a positive test with KCNS as found by Allmand & Webb.[5]

(3) *Stability of ferrous ions in the presence of air*

Sufficient ferric ammonium sulphate was added to aliquot portions of a 0·25% oxalic acid solution at pH 1·7 to give concentrations of 20, 30, 40 and 75 μg. of ferric iron/ml. Initially, nitrogen was passed through the solutions during irradiation; later, the nitrogen was replaced by air.

At intervals, 2-ml. samples were withdrawn and titrated with dye solution; the results are given in Table III, the ferric iron values being obtained by difference.

**Table III**

*Stability of ferrous ions on irradiation in the presence of air (dye titrations of 2-ml. aliquot portions of solution)*

| Experiment No. | | 1 | | | 2 | | | 3 | | | 4 | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| μg. of ferric iron/ml. | | 20 | | | 30 | | | 40 | | | 75 | | |
| Ratio | Wt. of iron / Wt. of oxalate | 1/125 | | | 1/83 | | | 1/62·5 | | | 1/33 | | |
| | ml. dye | μg. $Fe^2$ + found | μg. $Fe^3$ + calc. | ml. dye | μg. $Fe^2$ + found | μg. $Fe^3$ + calc. | ml. dye | μg. $Fe^2$ + found | μg. $Fe^3$ + calc. | ml. dye | μg. $Fe^2$ + found | μg. $Fe^3$ + calc. |
| 0 hours | 0 | 0 | 40 | 0 | 0 | 60 | 0 | 0 | 80 | 0 | 0 | 150 |
| | | | | | Nitrogen passing through the solutions during irradiation | | | | | | | |
| ½ hour | 0·55 | 44 | 0 | 0·80 | 64 | 0 | 1·10 | 88 | 0 | 2·0 | 160 | 0 |
| 1 ,, | 0·55 | 44 | 0 | 0·82 | 65 | 0 | 1·12 | 90 | 0 | 2·1 | 168 | 0 |
| | | | | | Air passing through the solutions during irradiation | | | | | | | |
| 2 hours | 0·05 | 4 | 36 | 0·65 | 52 | 8 | 1·00 | 80 | 0 | 2·1 | 168 | 0 |
| 2½ ,, | 0·0 | 0 | 40 | 0·50 | 40 | 20 | 0·95 | 76 | 4 | 2·0 | 160 | 0 |
| 3 ,, | 0·0 | 0 | 40 | 0·50 | 40 | 20 | 0·90 | 72 | 8 | 2·1 | 168 | 0 |

The results confirm that ferrous iron was formed on irradiation in an atmosphere of nitrogen; the behaviour of the ferrous iron subsequently on irradiation in air depended upon the ratio of iron to oxalate. When the ratio of weight of iron to weight of oxalate was 1 to 125, all the ferrous iron was reoxidized to the ferric state, whereas at a ratio of 1 to 33 none of the ferrous salt was reoxidized; at intermediate ratios some ferrous salt was reoxidized. This type of experiment was carried out many times using a greater range of ratios of iron to oxalate and the result was obtained that at a ratio of $\frac{\text{Weight of iron}}{\text{Weight of oxalate}} = \frac{1}{50}$ or greater, the iron remains in the ferrous state, whilst, at ratios of $\frac{1}{125}$ to $\frac{1}{250}$ or less, the iron remains in the ferric state.

In confirmation of this result it was found that ferrous salt was formed from ferric salt on irradiation in an atmosphere of air provided that the ratio was greater than $\frac{1}{50}$. This result is in harmony with the method adopted by Parker of determining ferrous iron with 1 : 10-phenanthroline as a measure of the photolysis of ferri-oxalate in which the ratio of weight of iron to weight of oxalate is 1 to 4·7, both in nitrogen and air. In the photochemical oxidation of ascorbic acid on irradiation of a standard iron solution in an atmosphere of air—where the ratio of weight of iron to weight of oxalate is 1 to 2500—it is believed that the ferrous iron formed is immediately reoxidized to ferric iron by the air.

## Discussion

The results of the experimental work are in accordance with the view that the first step in the photochemical oxidation of ascorbic acid (in oxalic acid solution in the presence of a trace of iron salt and with a stream of air bubbles passing through the solution) is the photochemical reduction of ferri-oxalate to the ferrous salt. Parker[9] suggests a mechanism based on a photo-sensitized electron-transfer reaction:

$$Fe^{3+}(C_2O_4)^{2-} \rightarrow Fe^{2+}(C_2O_4)^{-} \rightarrow Fe^{2+} + C_2O_4^{-}$$

The subsequent autoxidation of the ferrous ion proceeds according to Weiss[11] as follows:

$$Fe^{2+} + O_2 \rightleftharpoons Fe^{3+} + O_2^-$$

the complexing with oxalate favouring an equilibrium shift to the right. It is suggested that the oxidation of the ascorbic acid results from the activity of the $HO_2$ and OH radicals which are produced in solution (cf. Posner,[12] Lu Valle & Weissberger[13]).

## Summary

(1) When a solution at pH 1·7 containing 0·25% oxalic acid, 0·02% ascorbic acid and ferric ammonium sulphate to give a concentration of 11·6 μg. of ferric iron per ml. was irradiated while a stream of nitrogen was passed through the solution, it developed additional reducing power as measured by the dye solution. This observation is in accordance with the view that the initial step in the photochemical oxidation of ascorbic acid is a photochemical reduction of ferri-oxalate to ferrous salt.

(2) Although ferric salts are known to oxidize ascorbic acid in sulphuric acid solution, it has been shown that they do not do so in oxalic acid solution. It is therefore suggested that the oxidation of ascorbic acid is dependent upon OH and $HO_2$ radicals which are produced in solution as a result of the autoxidation of the ferrous ions derived from the photochemical reduction of the ferri-oxalate.

(3) When aliquot portions of 0·25% oxalic acid solution at pH 1·7 containing ferric ammonium sulphate to give concentrations of (i) 20, (ii) 30, (iii) 40 and (iv) 75 μg. of ferric iron were irradiated in an atmosphere of nitrogen, reducing power (towards the dye) developed consistent with complete reduction to ferrous ions. Replacement of the nitrogen by air resulted in complete reoxidation to ferric salt in solution (i) and no reoxidation in solution (iv); partial reoxidation occurred in solutions (ii) and (iii). With air passing through the solution, the oxidation level of the iron therefore depends upon the ratio of iron to oxalate; the greater the proportion of iron, the greater is the tendency to remain in the reduced form. Ferrous ions are actually formed from ferric ions in oxalic acid solution in an atmosphere of air when the ratio of iron to oxalate is $>$ 1 to 50.

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# STUDIES ON THE NITROGEN METABOLISM OF THE ENSILAGE PROCESS

By A. R. KEMBLE*

The overall changes in nitrogen distribution occurring in laboratory silage have been investigated and, in particular, quantitative determinations of most of the monoamino-monocarboxylic acids have been made. Both good and bad silages have been studied. In the good silage the amounts of free amino-acids present were, in varying degrees, less than those calculated from the extent of protein breakdown which had occurred. In the bad silage, a considerable excess of alanine above that which could have been formed by uniform proteolysis was present during the first three weeks of the fermentation, and, after eight weeks, α-aminobutyric acid began to appear.

When some micro-organism-free grass was ' ensiled ', extensive proteolysis occurred, excess alanine was again formed but no free ammonia appeared. These results enable a distinction to be drawn between some of the enzymic and bacterial reactions which occur simultaneously in the silo.

## Introduction

One of the potentialities of the ensilage process is the possibility of preserving young high-protein fodder for winter use. Consequently the course and extent of the proteolysis which commences immediately after cutting the fodder has always been of particular interest. The problem of estimating the individual amino-acids formed has long been a matter of some difficulty, but the advent of chromatography did not immediately solve the problem because of the adverse effect exerted by the many salts which are an integral part of any plant extract. However, a method has been evolved which allows a relatively rapid determination of the majority of the monoamino-monocarboxylic acids to be made[1] and this method has been applied in the present work.

The problem of deciding which products of the silage fermentation are attributable to plant enzymes and which to bacterial processes has also been the subject of much discussion.[2] It is no easy matter to differentiate between these two effects. Therefore the provision of a small quantity of microbe-free grass by Dr. Stirling of the Edinburgh and East of Scotland College of Agriculture was of particular value in allowing preliminary work on this problem to be carried out.

## Experimental

### *Analyses*

Total nitrogen, soluble nitrogen, volatile base, total amide, glutamine, α-amino-acid nitrogen, alcohol-insoluble peptide nitrogen and monoamino-monocarboxylic acids were all determined as have been described previously.[3]

α-Aminobutyric acid was found partially to overlap proline on the *iso*butanol–methyl ethyl ketone–water chromatogram, but it was not formed in silage until proline had disappeared. Thus it could be estimated after elution from a chromatogram run in the above solvent system. Recovery factor[1] = 1·004.

### *Preparation of experimental silos*

Young perennial rye-grass S.24, hay strain, was cut on 14 June, 1954. The grass was 3–6 in. in height, moisture content 85·9%, total nitrogen 4·71% (on dry matter).

The cut grass was thoroughly mixed and a sample (250 g.) was immediately extracted with boiling water.

Another sample (4 kg.) was taken and thoroughly mixed with a washed suspension of anaerobic bacteria in water (138 ml.). Portions of this material were tightly and, as far as possible, uniformly packed into six sterilized quart milk bottles. The contents of the bottles ranged from 483·8 g. to 501·0 g. in weight. The bottles were stoppered with rubber bungs fitted

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with mercury seals which allowed gas to escape although preventing the entrance of air and airborne bacteria. These silos were placed in an incubator kept at 30°. A bottle was opened and a portion of its contents (approx. 300 g.) extracted with boiling water after 1, 3, 7, 21, 57 and 147 days. These silos will be referred to as the A series.

A third portion (3·5 kg.) of the cut grass was taken and mixed thoroughly while a washed suspension (25 ml.) of lactobacilli was sprinkled on to it. From this material six samples (500 g.) were taken. Each sample was thoroughly mixed with glucose solution (12·5 g./19 ml. water) and packed into quart milk bottles. These silos (the L series) were sealed, stored and examined as described above.

*Preparation of micro-organism-free silage*

A sample (8·2 g.) of young timothy grass (3–4 in. in height) from the sterile growth chamber [4] was cut, packed into a test-tube and stoppered with a mercury seal before removal. The test-tube was kept at 30° for 16 days before a portion of the contents (7·63 g.) was extracted with boiling water. The remainder was declared microbe-free after examination in the Bacteriology Department of the Edinburgh and East of Scotland College of Agriculture.

## Results

The results for the ensilage experiments, other than the amino-acid determinations, are contained in Table I (Series L) and Table II (Series A). In Tables III (Series L) and IV (Series A), the determinations of the individual amino-acids are compared with the maximum values possible, assuming uniform protein breakdown. These values have been calculated from the expression, $A_f + A_p\left(\frac{S_n - S_f}{100}\right)$, where $A_f$ is the free amino-acid content of fresh grass, $A_p$ is the amino-acid content of the protein and $S_f$ and $S_n$ are the soluble nitrogen fractions (% of total N) of the fresh grass and sample taken on the $n$th day, respectively.

### Table I

*Changes in pH and nitrogen distribution in Series L*

| | Days | | | | | | |
|---|---|---|---|---|---|---|---|
| | 0 | 1 | 3 | 7 | 21 | 57 | 147 |
| pH | 6·1 | 4·2 | 3·9 | 3·9 | 3·9 | 3·8 | 3·9 |
| | % of total N | | | | | | |
| Soluble N | 18·2 | 37·6 | 46·9 | 54·0 | 54·8 | 57·7 | 59·2 |
| Volatile base N | 0·5 | 1·7 | 2·2 | 2·3 | 2·4 | 2·6 | 3·0 |
| Glutamine-N | 3·0 | 0·8 | 0·5 | 0·9 | 0·5 | 0·6 | 0·1 |
| Asparagine-N | 2·0 | 2·5 | 3·5 | 3·8 | 4·5 | 4·8 | 2·1 |
| α-Amino-acid-N (less ½ of amide-N) | 5·0 | 10·0 | 11·8 | 15·2 | 16·8 | 18·4 | 20·6 |
| Alcohol-insoluble peptide-N | 1·7 | 1·2 | 1·1 | 1·6 | 1·5 | 1·8 | 1·9 |

### Table II

*Changes in pH and nitrogen distribution in Series A*

| | Days | | | | | | |
|---|---|---|---|---|---|---|---|
| | 0 | 1 | 3 | 7 | 21 | 57 | 147 |
| pH | 6·1 | 6·2 | 5·3 | 5·6 | 6·7 | 7·2 | 7·3 |
| | % of total N | | | | | | |
| Soluble N | 18·2 | 42·3 | 63·2 | 63·8 | 70 | 83·6 | 75·3 |
| Water-insoluble N | — | — | — | — | — | — | 22·8 |
| Volatile base N | 0·5 | 3·1 | 5·2 | 7·3 | 17·0 | 55·9 | 55·8 |
| Glutamine-N | 3·0 | 1·2 | 2·7 | 1·6 | 0·8 | 0 | 0 |
| Asparagine-N | 2·0 | 2·4 | 2·3 | 1·6 | 1·4 | 0 | 0 |
| α-Amino-acid-N (less ½ of amide N) | 5·0 | 11·5 | 19·3 | 21·8 | 20·6 | 12·2 | 6·6 |
| Alcohol-insoluble peptide-N | 1·7 | 2·3 | 1·5 | 1·0 | 0·9 | 0·5 | 0·1 |

**Table III**

*Monoamino-monocarboxylic acid content of Series L silage. (All values are N as % of total nitrogen)*

| | | Days | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | 0 | 1 | 3 | 7 | 21 | 57 | 147 |
| Glycine | Found | 0·08 | 0·47 | 0·69 | 0·88 | 0·88 | 1·12 | 1·32 |
| | Expected | — | 1·40 | 2·03 | 2·51 | 2·57 | 2·78 | 2·87 |
| Serine | Found | 0·40 | 0·67 | 0·59 | 0·82 | 0·94 | 1·21 | 1·30 |
| | Expected | — | 1·20 | 1·58 | 1·87 | 1·90 | 2·02 | 2·08 |
| Threonine | Found | 0·26 | 0·45 | 0·67 | 0·85 | 0·96 | 1·17 | 1·24 |
| | Expected | — | 1·09 | 1·49 | 1·80 | 1·83 | 1·96 | 2·02 |
| Alanine | Found | 0·27 | 1·52 | 1·65 | 2·09 | 2·27 | 2·36 | 2·91 |
| | Expected | — | 1·39 | 1·93 | 2·35 | 2·39 | 2·56 | 2·65 |
| Proline | Found | 0·11 | 0·50 | 0·58 | 0·70 | 1·01 | 1·22 | 1·24 |
| | Expected | — | 0·79 | 1·11 | 1·36 | 1·39 | 1·49 | 1·55 |
| Tyrosine | Found | 0·05 | 0·26 | 0·40 | 0·51 | 0·56 | 0·57 | 0·64 |
| | Expected | — | 0·48 | 0·68 | 0·84 | 0·86 | 0·92 | 0·95 |
| Valine | Found | 0·08 | 0·54 | 0·72 | 0·91 | 1·10 | 1·17 | 1·39 |
| | Expected | — | 0·97 | 1·40 | 1·73 | 1·76 | 1·90 | 1·97 |
| Methionine | Found | 0·02 | 0·17 | 0·23 | 0·28 | 0·33 | 0·36 | 0·39 |
| | Expected | — | 0·25 | 0·36 | 0·45 | 0·46 | 0·49 | 0·51 |
| Phenylalanine | Found | 0·05 | 0·49 | 0·53 | 0·71 | 0·82 | 0·96 | 1·11 |
| | Expected | — | 0·73 | 1·05 | 1·30 | 1·33 | 1·43 | 1·49 |
| *iso*Leucine | Found | 0·03 | 0·33 | 0·43 | 0·59 | 0·64 | 0·64 | 0·87 |
| | Expected | — | 0·69 | 1·01 | 1·25 | 1·27 | 1·37 | 1·42 |
| Leucine | Found | 0·05 | 0·78 | 1·08 | 1·36 | 1·76 | 1·89 | 2·11 |
| | Expected | — | 1·14 | 1·66 | 2·06 | 2·10 | 2·26 | 2·35 |
| Tryptophan | No free amino-acid detected | | | | | | | |

With the exception of tyrosine (2·4%) and methionine (1·5%),[5] the values for the amino-acid content of the protein have been taken from a recent publication by Kemble & Macpherson.[6] Evidence in favour of the assumption that protein breakdown during ensilage is uniform is also presented in this paper.

The results for the micro-organism-free experiment are given in Table V. Because of the small quantity of material available, the number of determinations was limited.

## Discussion

The immediate objective of the experiments was to make good and bad silage from the same grass. Thus comparisons between the two fermentations would not be complicated by differences in starting material. A young high-protein (crude protein 29·4%) grass was used because such material is notoriously difficult to ensile and yet, when well preserved, provides a most valuable winter fodder.

In order that the experiment should have every chance of success, a suspension of anaerobic bacteria (Clostridia) known to be present in putrefied silage was added to Series A, while a suspension of lactobacilli together with a nutrient (glucose) was added to Series L.

These treatments were successful in producing the desired results. In Series A, the available sugar was exhausted before sufficient acidity for preservation had been generated. Thus the proteolytic enzymes and anaerobic bacteria continued to function, resulting in a putrefied product of high pH and high volatile base content (Tables II and VI).

In Series L, the added glucose was sufficient to enable the lactobacilli to produce the acidity necessary for preservation (Tables I and VI). Even so, approximately one-third of the protein

## Table IV

*Monoamino-monocarboxylic acid content of Series A silage. (All values are N as % of total nitrogen)*

| | | Days | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | 0 | 1 | 3 | 7 | 21 | 57 | 147 |
| Glycine | Found | 0·08 | 0·76 | 1·94 | 2·14 | 3·01 | 0·79 | 0·16 |
| | Expected | — | 1·72 | 3·14 | 3·18 | 3·67 | 4·53 | 3·96 |
| Serine | Found | 0·40 | 0·95 | 1·51 | 1·41 | 0·55 | 0 | 0 |
| | Expected | — | 1·39 | 2·24 | 2·27 | 2·56 | 3·08 | 2·74 |
| Threonine | Found | 0·26 | 1·00 | 1·55 | 2·15 | 1·53 | 0·32 | 0 |
| | Expected | — | 1·30 | 2·20 | 2·22 | 2·53 | 3·07 | 2·72 |
| Alanine | Found | 0·27 | 2·18 | 3·78 | 3·76 | 4·31 | 1·52 | 1·31 |
| | Expected | — | 1·67 | 2·88 | 2·91 | 3·33 | 4·06 | 3·58 |
| Proline | Found | 0·11 | 0·66 | 0·99 | 1·42 | 1·56 | 0·44 | 0 |
| | Expected | — | 0·95 | 1·69 | 1·71 | 1·95 | 2·40 | 2·11 |
| α-Aminobutyric acid | Found | 0 | 0 | 0 | 0 | 0 | trace | 0·27 |
| | Expected | — | 0 | 0 | 0 | 0 | 0 | 0 |
| Tyrosine | Found | 0·05 | 0·46 | 0·92 | 0·70 | 0·61 | 0·29 | 0 |
| | Expected | — | 0·58 | 1·04 | 1·05 | 1·21 | 1·49 | 1·31 |
| Valine | Found | 0·08 | 1·01 | 1·93 | 2·25 | 2·40 | 0·99 | 1·06 |
| | Expected | — | 1·19 | 2·15 | 2·18 | 2·51 | 3·09 | 2·71 |
| Methionine | Found | 0·02 | 0·23 | 0·28 | 0·29 | 0·43 | 0·07 | 0·04 |
| | Expected | — | 0·31 | 0·56 | 0·57 | 0·65 | 0·80 | 0·71 |
| Phenylalanine | Found | 0·05 | 0·65 | 1·18 | 1·25 | 1·48 | 0·54 | 0·31 |
| | Expected | — | 0·89 | 1·63 | 1·65 | 1·89 | 2·34 | 2·05 |
| *iso*Leucine | Found | 0·03 | 0·56 | 1·35 | 1·55 | 1·66 | 0·62 | 0·51 |
| | Expected | — | 0·85 | 1·56 | 1·58 | 1·83 | 2·25 | 1·97 |
| Leucine | Found | 0·05 | 1·11 | 2·09 | 2·36 | 2·31 | 1·03 | 0·92 |
| | Expected | — | 1·39 | 2·57 | 2·60 | 3·01 | 3·71 | 3·25 |
| Tryptophan | No free amino-acid detected | | | | | | | |

## Table V

*Nitrogen distribution and pH of micro-organism-free grass 16 days after 'ensilage'*

| | Found | Expected |
|---|---|---|
| pH | 6·1 | — |
| | % of total N | % of total N |
| Soluble N | 59·9 | — |
| N as volatile base | 1·6 | — |
| Glycine | 1·9 | 3·2 |
| Serine | 1·9 | 2·2 |
| Threonine | 2·3 | 2·1 |
| Alanine | 4·2 | 2·9 |
| Proline | 1·6 | 1·8 |
| Tyrosine | 0·7 | 1·1 |
| Valine | 2·1 | 2·2 |
| Methionine | 0·4 | 0·6 |
| Phenylalanine | 1·3 | 1·7 |
| *iso*Leucine | 1·4 | 1·6 |
| Leucine | 2·6 | 2·7 |

had been degraded within three days of ensiling, i.e. before the inhibitory action of a low pH had become effective. When mineral acid was used to produce silage of a pH below 4, further proteolysis was prevented.[7] In the present work, however, a slow but steady increase in soluble nitrogen was observed at pH 3·8 although there was no appreciable increase in volatile base (Table I). Thus four months after ensiling, a product was obtained which had the pH, volatile

**Table VI**

*Glucose + fructose + sucrose + fructosan content of ensiled materials (% dry matter)* *

| | Days | | | |
|---|---|---|---|---|
| | 0 | 1 | 3 | 7 |
| Series A | 1·31 | 0·64 | 0·25 | 0·02 |
| Series L | 3·81 | 1·71 | 0·48 | 0·08 |

* Information kindly supplied prior to publication by Mr. D. Mackenzie

base content and odour of good silage even though half of the original protein had been degraded. The extent to which a similar breakdown would occur in field silos would depend on the attainment and duration of a temperature of 30°.

Determination of the nitrogen remaining in the residue after water extraction showed that the reduction in soluble nitrogen content (Series A) between the 57th and 147th days was not due to a loss of nitrogen from the system (Table II). During a complex chemical reaction lasting for four months, any slight differences between the initial contents of two bottles would be magnified, and this is the probable explanation of the anomalous result.

When grass is allowed to wilt, almost all the nitrogen rendered soluble by proteolysis can be recovered as alcohol-insoluble nitrogen, amide, amino-acid or volatile base.[3] This is certainly not true for the silage fermentations where, after seven days, only 67% (Series L) and 73% (Series A) of the degraded protein nitrogen reappeared in these forms. Considerable amounts of $\gamma$-aminobutyric acid are formed during ensilage but it is unlikely that this could account for all the missing nitrogen. Recent work by Ferguson & Terry[8] has shown that in some samples of herbage only a relatively small proportion of the peptides are precipitated from an aqueous extract by the addition of three volumes of alcohol. During the present investigation, the fraction of the aqueous extract of Series A silage (7 days) soluble in 75% alcohol was hydrolysed with 6N-hydrochloric acid. No increase in $\alpha$-amino-acid content was found. These conditions of hydrolysis are known to cause some decomposition of amino-acids but the result shows that very little of the missing 30% total N can be in the form of alcohol-soluble peptide.

Determination of the monoamino-monocarboxylic acids showed that all the amino-acids, with the exception of alanine in Series A, were present in amounts less than those calculated from protein breakdown. Previous work has shown that the protein remaining after a silage fermentation is little different in monoamino-acid composition from the original grass protein.[6] Thus the observed deficiencies in amino-acid content will be a measure of the rate at which the amino-acids are metabolized after liberation.

In Series L (Table III) there was a rapid formation of amino-acids during the first three days, followed by a slow but steady rise during the ensuing four months. In Series A (Table IV) there was a similar initial rapid rise but there came a stage for each amino-acid when its degradation was more rapid than its release by proteolysis. This time varied from three days (tyrosine) to three weeks (glycine, alanine and phenylalanine).

After 57 days the water extract of Series A contained a new substance giving a faint spot on a qualitative chromatogram sprayed with ninhydrin. After 147 days the spot was more intense. It corresponded chromatographically in three different solvents with $\alpha$-aminobutyric acid which has been reported by Robertson & Barnett[9] to appear after eight weeks in anaerobic slurries of kale. These authors suggest that $\alpha$-aminobutyric acid might be formed from threonine by coliform organisms. No guidance on the matter could be gained from the present work because all the amino-acids had been extensively degraded.

During the first three weeks of the Series A fermentation, alanine was found to be present considerably in excess of the calculated amount (Table IV). This suggests that some synthesis of alanine by the anaerobic bacteria had occurred. However, a similar excess was found in ' micro-organism-free silage ' (Table V). Thus the excess alanine must have been formed by plant enzymes. Presumably these enzymes were inhibited in Series L by the acidity of the substrate, a hypothesis which receives some support from the observation that a slight excess of alanine occurred in Series L during the first day of the fermentation, i.e. before the pH had dropped to its final low level (Table III).

It is a matter of some interest that, in cut grass, excess alanine is formed under moist anaerobic conditions, excess asparagine under moist aerobic conditions[3] and excess proline during aerobic wilting.[3]

The experiment with micro-organism-free grass supports the general view that formation of ammonia from amino-acids during ensilage is a function of the anaerobic bacteria present rather than of the plant enzymes. Evidence to the contrary, however, has been presented.[10]

Furthermore, the use of micro-organism-free grass shows that the rapid proteolysis at the commencement of the ensilage process is caused by plant enzymes. It also shows that these enzymes do not produce substances which change the pH from that of the fresh grass.

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# SOME SEED FATS OF THE *SANTALACEAE* FAMILY

By H. H. HATT and R. SCHOENFELD

The octadecenynoic acid, ximenynic acid, is present in quantity in the seed fats of three members of the *Santalum* genus: *S. acuminatum*, *S. Murrayanum* and *S. spicatum*. The composition of the fatty acids from the seed fat of *S. acuminatum* was calculated to be: palmitic acid, 3%; stearic acid, 1%; oleic acid, 50%; linolenic acid, 2%; and ximenynic acid, 44%. The higher ethenoid acids which accompany ximenynic acid in the seed fats of the *Ximenia* genus, are here either absent, or present only in small amount.

## Introduction

The conjugated octadecenynoic acid known as ximenynic acid was first identified by South African workers[1, 2] in the seed fats of three members of the *Ximenia* genus. There it was present together with much oleic acid and a series of high molecular weight monoethenoid acids which were then found for the first time in seed fats.[3] Subsequently in this laboratory[4] ximenynic acid was shown to be present in the seed fats of some Australian species of *Santalum*; in fact, the proportion there is so high that these fats are a convenient natural source of the acid, which can be isolated in the manner already described.[4]

In reporting the presence of ximenynic acid in these *Santalum* seed fats, the suggestion was made that the 'santalbic acid' of unknown structure found[5] in the seed fat of the closely related

*Santalum album* of India was probably identical with ximenynic acid. This suggestion was almost at once proved true by Gunstone,[6] who working independently showed the identity of santalbic acid with ximenynic acid. The acid has also been synthesized.[7]

This acid is thus present in the fats of two closely related families, for the *Ximenia* genus belongs to the *Olacaceae* and the *Santalum* genus to the *Santalaceae*. It was therefore of interest to discover whether the series of high molecular weight ethenoid acids were also present in the *Santalum* seed fats.

Three seed fats were examined: *Santalum acuminatum* (syn. *Fusanus acuminatus*), the sweet quandong; *Santalum Murrayanum* (syn. *F. persicarius*), the bitter quandong; *Santalum spicatum* (syn. *F. spicatus* or *Eucarya spicata*), the Australian sandalwood. Only the fat of the sweet quandong was examined in detail, but with the others sufficient was done to show that the components present were principally those found in the sweet quandong fat.

## Experimental

*Santalum acuminatum* (sweet quandong)

Samples of the ripe nuts were obtained from Mildura and Wail in Victoria and from Comet Vale in Western Australia. The seeds varied considerably in size, and had an average weight of 2 g. They were very hard and were cracked in a small vice. The kernels formed, on average, 17% of the whole nut and had a moisture content of 3·3%.

The presence of a rubber-like material in the kernels made extraction of the fat difficult. Best results were obtained by grinding coarsely in a coffee mill, extracting twice with cold light petroleum and then transferring the residue to a Soxhlet and extracting exhaustively with the same solvent. A suspension in the extract was allowed to settle and removed by decantation and washing. After removal of the solvent from the combined extracts, two volumes of acetone were added, which caused suspended matter and 'rubber' to coagulate; these were filtered off using filter aid and the fat obtained from the filtrate. All operations were carried out under nitrogen. The yields of fat from the three samples were 66% (Mildura), 57% (Wail) and 55% (Western Australia). These figures refer to moisture-free kernels. The rubber-like resin formed about 2% of the kernel.

A typical seed fat from the Mildura sample was an optically inactive pale yellow oil with the following physical and chemical properties:

$d_4^{25}$ 0·9231 $n_D^{25}$ 1·4778
Acid value 0·8
Saponification value 193·4
Glycerol yield 10·1%
Iodine value (Wijs) 94·5–99
(variable according to time)
Reichert Meissl number 0·55
Polenske number 0·35
Yield of fatty acids 94·3%
Unsaponifiable matter 1·0%
Total saturated acids (Bertram) 4·2%.

At 229 m$\mu$. the fat had a specific extinction coefficient of $k = 22{\cdot}6$. For pure methyl ximenynate the value for $k$ is 54·9, whence the proportion of the acid in the seed fat is 41·2% and in the total acids 43·7%.

A sample of the fatty acids obtained by cold saponification was methylated with diazomethane and hydrogenated in alcohol over Raney nickel at 110–120° under pressure. 5·26 g. of the saturated methyl esters so obtained (saponification equivalent 297·6; iodine value 0) were submitted to amplified distillation with 80 g. of carrier oil using a spinning band column at a pressure of 1·0 mm. Hg. The composition of these methyl esters was then determined as previously described[8] and found to be:

| | |
|---|---|
| $C_{16}$ | 3·5% |
| $C_{18}$ | 91·3% |
| $C_{20}$ | traces |
| Higher boiling | 1·3% |
| Undistillable | 3·4% |

The higher boiling and undistillable portions were separated from the carrier oil but were not examined in detail. They did not appear to be normal saturated acids and were considered likely to be derived from polymerization of ximenynic acid and accompanying break-down products.

A sample (41·9 g.) of the methyl esters of the fatty acids was fractionally distilled in a spinning band column at 1·00 mm. pressure with the use of 30 g. of high-boiling mineral oil as a chaser. As the distillation graph (Fig. 1) shows, a very sharp separation was obtained between methyl ximenynate (b.p. 169°) and the other $C_{18}$ esters. Spectrophotometric assay of the fractions from the lower $C_{18}$ plateau showed that only 0·6% of methyl ximenynate was present. The lower plateau fractions had iodine values of 94·2 and consisted mainly of oleic acid. The acids from this lower plateau were oxidized with alkaline permanganate and the presence of oleic acid was established by isolation of 9 : 10-dihydroxyoctadecanoic acid, m.p. 129–129·5°, unchanged by admixture with an authentic specimen. No tetrahydroxyoctadecanoic acid was found in the products of oxidation, so that linoleic acid can have been present only in traces.

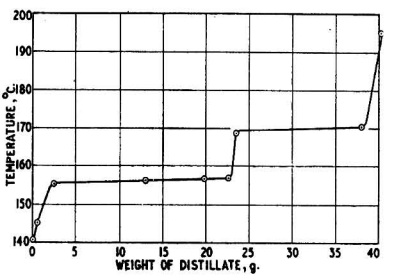


FIG. 1.—Santalum acuminatum *distillation of methyl esters* (41·9 *g.*) *at* 1·00 *mm.*

However, bromination of the acids from this plateau revealed the presence of linolenic acid since appreciable amounts of a crystalline bromide were obtained, which after repeated crystallization melted at 181–181·5° but still darkened during melting and left a small amount of insoluble material. A mixed melting point with 9 : 10 : 12 : 13 : 15 : 16-hexabromo-octadecanoic acid showed no depression, but the bromine content was somewhat low (Found : Br, 61·3. Calc. for $C_{18}H_{30}Br_6O_2$ 63·3%).

Of the other acids, palmitic acid was isolated from the first fraction of the simple distillation and identified by its melting point (63°) and that of its methyl ester (30°).

The composition of the fatty acids from this fat was calculated using the extinction coefficient at 229 mμ., the content of saturated acids (Bertram) and the results of the amplified and simple distillations. The iodine values of distillation fractions were determined and in using these it was assumed that linoleic acid was not present. The composition was then found to be :

| | |
|---|---|
| Palmitic acid | 3% |
| Stearic acid | 1% |
| Oleic acid | 50% |
| Linolenic acid | 2% |
| Ximenynic acid | 44% |

*Santalum Murrayanum* (bitter quandong)

The sample of seeds (nuts) was obtained from Wail, Victoria. It was examined in the manner described for the sweet quandong. The data obtained are shown in Table I.

Bromination of the fatty acids in ether gave a small amount of bromo-acids, which after repeated crystallization from benzene melted at 181°. The presence of oleic acid was also established by oxidation to 9 : 10-dihydroxyoctadecanoic acid, m.p. 129–129·5°.

*Santalum spicatum* (Australian sandalwood)

This tree is the source of commercial sandalwood and sandalwood oil. It is confined to Western Australia. The seeds (nuts) were examined in the manner described for the sweet quandong and the data obtained shown in Table I.

Fractional distillation of the methyl esters at 1 mm. pressure, in the manner described for the sweet quandong esters, gave a similar distillation curve. The $C_{18}$ and ximenynic ester plateaux were again the principal features, and in conformity with the lower amount of ximenynic ester present in this fat, its fraction was correspondingly smaller and was compensated by an increase in the $C_{18}$ fraction. The $C_{16}$ fraction was again small and no certain higher fraction was obtained.

**Table I**

*Composition of seeds and seed fats*

| | *Santalum Murrayanum* | *Santalum spicatum* |
|---|---|---|
| Properties of seed | | |
| Average wt. of nuts | 2·4 g. (range 1·5–3·8 g.) | 2·2 g. (range 1·0–5·0 g.) |
| Yield of kernels | 19·0% | 28·3% |
| Moisture content of kernels | 4·3% | 3·2% |
| Fat content of kernels (moisture free) | 71·4% | 61·5% |
| Resin content | 2·2% | Present but not estimated |
| N in oil-free kernels | 7·8% | — |
| (calc. protein | 49%) | |
| Ash from oil-free kernels | 5·14% | — |
| Properties of seed fat | | |
| Colour | pale yellow | pale yellow |
| $[\alpha]_D^{25}$ | − 0·26% | — |
| $n_D^{25}$ | 1·4783 | 1·4752 |
| Saponification value | 189·3 | 193·3 |
| Iodine value (Wijs, 1 h.) | 97·7 | 86·5 |
| Acetyl value | — | 0·0% |
| Glycerol yield | 9·6% | 9·4% |
| Unsaponifiable matter | 1·0% | 0·5% |
| Yield of fatty acids | 94·4% | 94·3% |
| Ximenynic acid in fatty acids | 45·0% | 36·3% |
| Saturated acids in fatty acids (Bertram) | 4·3% | — |

## Discussion

Including *Santalum album*, four seed fats of the *Santalum* genus have now been shown to contain ximenynic acid in quantity and it is probable that the presence of this acid in the seed fat will prove to be a characteristic of all members of this genus. There is a notable difference between these fats and those of the *Ximenia* genus. Both contain ximenynic acid in quantity, but, unlike *Ximenia*, in the *Santalum* genus it is not accompanied by high molecular weight ethenoid acids. The proportion of these acids, if present at all in *Santalum* fats, can only be very low.

It is of interest that, in the *Santalum* fats, oleic acid is present in large proportion together with significant amounts of linolenic acid, but that linoleic acid is present only in small amount or absent. Hilditch[9] has suggested the general biosynthetic sequence : linolenic, linoleic, oleic acid, and it may be that in the *Santalum* genus ximenynic acid acts as an intermediate between linolenic and oleic acids, opening a route which avoids linoleic acid. The occurrence together of linolenic and ximenynic acids suggests they have a common precursor.

## Acknowledgments

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# CARBON-NITROGEN RELATIONSHIPS IN SOIL. I.—The Immobilization of Nitrogen in the Presence of Carbon Compounds

By G. W. WINSOR and A. G. POLLARD

Immobilization of nitrogen has been studied in soils treated with starch and sucrose. The conversion of inorganic nitrogen into organic form reached its maximum on incubation at 23·5° for approximately two days. The organisms concerned in the immobilization of nitrogen showed a marked preference for ammonia rather than nitrate, although some nitrate was invariably assimilated at the higher levels of added carbon compounds. The assimilation of ammonia lowered the pH of the soil, whereas assimilation of nitrate increased it markedly.

## Introduction

When an organic material which can be decomposed by micro-organisms is added to the soil, a complex series of reactions results. The heterotrophic organisms of the soil use the organic material as a source of energy, and a rapid increase occurs in their numbers. Part of the added carbon is evolved as carbon dioxide, while part is used by the organisms in the synthesis of their own cell substance. This synthesis of microbial tissue is accompanied by the assimilation of nitrogen and various other elements by the micro-organisms. As the supply of organic material becomes exhausted, the population of soil organisms declines, and the mineralization of microbial tissue predominates over its formation.

The need to maintain the content of organic matter in cultivated soils has led to the application of a wide range of carbonaceous materials, and it has long been known that such treatments can induce a temporary deficiency of inorganic nitrogen available for plant growth. The literature of this subject is too extensive to be reviewed here in detail, but examples include investigations of the effects of straw,[1, 2] sawdust,[3, 4] molasses[5] and other materials. The nitrogen content of an organic material can itself be used as a rough guide to its effect on the inorganic nitrogen of the soil, materials of high nitrogen content causing accumulation of ammonia and nitrate, whereas materials containing little or no nitrogen lead to immobilization of the inorganic nitrogen already present in the soil.[6, 7]

More precise information concerning the effect of organic materials on the levels of inorganic nitrogen in the soil may be based on their carbon/nitrogen ratios ; the quantitative aspect of this relationship will be reviewed in a subsequent paper. Studies of the effect of carbon/nitrogen ratio on nitrogen transformations in the soil have been made with fungal and bacterial tissue,[8, 9] organic fertilizers[10] and amino-acids.[11, 12] There is also an extensive literature concerning the decomposition of cellulose in relation to the nitrogen supply, of which the work of Waksman & Skinner[13] may be cited as an example.

The soil perfusion technique has been used by Lees[14] to study immobilization of nitrate-nitrogen in the presence of organic compounds. With glucose and sucrose, immobilization of nitrogen reached its maximum in two to four days, after which the nitrate concentration rose again. The amounts of nitrogen immobilized were found to increase as the ratio of soil to solution in the perfusion units was decreased.

The relationship between carbon/nitrogen ratio and nitrogen transformations in the soil is influenced by the ease of decomposition of the various constituents present,[9, 10] and our knowledge of the decomposition of complex materials in the soil is limited by the analytical procedures available. The present investigations were therefore made with relatively simple materials in order to give further information concerning the immobilization of nitrogen in the soil. Apart from certain preliminary experiments with starch, sucrose was used as the source of carbon, while nitrogen was supplied in the form of ammonium salts and nitrates. The amounts of nitrogen immobilized have been calculated as the difference in content of inorganic nitrogen between soil treated with inorganic nitrogen in the presence and absence of an added carbon compound. The term ' control soil ' refers to soil receiving no treatment other than the maintenance of moisture content.

Apart from transformations of nitrogen in the soil resulting from the processes of mineralization and immobilization, small amounts of nitrogen may also be gained by fixation or lost in the form of ammonia or gaseous nitrogen. The inadequacy of the Kjeldahl procedure for the evaluation of small changes in the total nitrogen content of soils has been stressed by Broadbent & Stojanovic,[14a] and no attempt has been made to study changes in total nitrogen content in the present investigation. The possibility of small changes in the nitrogen content of the soils cannot therefore be entirely excluded, but considerable evidence is available to show that in the present experiments the primary effects are due to conversion of nitrogen from inorganic to organic forms in the soil rather than to losses of nitrogen from the system.

## Experimental methods

In the preliminary experiments, 1·5 kg. batches of moist soil were treated with starch and ammonium salts in solid form, and stored in glazed earthenware pots. The pots were covered with glass plates to reduce moisture losses. For the determination of ammonia and nitrate, the contents of the pots were turned out and thoroughly mixed, 50–100 g. samples of soil being removed for analysis. Further samples were used in the determination of moisture content.

In subsequent experiments, flasks containing 50–100 g. of soil were used, these being lightly plugged with cotton wool. For the determination of ammonia and nitrate the entire contents of duplicate flasks were extracted. The weights of the flasks containing soil were checked at intervals, and any losses of moisture made good. Solutions containing sugar and inorganic nitrogen were applied to the soil with calibrated 3- or 5-ml. pipettes. The moisture content of the soil was so adjusted before pipetting that the added solution raised it to 90% of the moisture equivalent as determined by the suction method of Bouyoucos.[15] All soils under investigation were incubated in darkness at 23·5°.

For the estimation of ammonia and nitrate, 50–100-g. samples of moist soil were extracted with approximately 600 ml. of N-sodium chloride. Ammonia was determined in the filtrate by addition of freshly-ignited magnesia and distillation into 0·02N-acid. For the determination of nitrate in the same soil extract, distillation was continued after addition of 3 g. of Devarda's alloy.

Phosphate was determined colorimetrically in 0·5N-acetic acid extracts, using the molybdenum blue procedure. All results are expressed on the basis of oven-dried soil.

Measurements of pH were made with a glass electrode in soil suspensions shaken for one hour. Except where otherwise stated, the soil suspensions were prepared at a water/soil ratio of 2·5 : 1.

## Results

*The effect of mixtures of starch and ammonium salts upon the levels of inorganic nitrogen in the soil*

In a preliminary experiment, pots containing 1·5 kg. of a market-garden soil were treated with di-ammonium hydrogen phosphate in quantities corresponding to 300 p.p.m. of added nitrogen. Additional treatments with 0·15, 0·25 and 0·35% starch were made in duplicate. The carbon content of the starch used was determined by combustion to be 37·4%, the carbon/nitrogen ratios of the added mixtures thus being 1·87, 3·12 and 4·36, respectively. A further quantity of soil was left as a control, receiving no treatment other than the maintenance of moisture content. Analyses for ammonia and nitrate were made until nitrification was complete. The results are shown as differences from corresponding values for the control soil in Fig. 1.

In the early stages of the experiment, very wide differences were found in the ammonia content of soil treated at the different carbon/nitrogen ratios, the levels of ammonia at any one time being in inverse order to the carbon/nitrogen ratios of the added materials. In contrast, however, no significant differences were found between the amounts of nitrate formed at the different carbon/nitrogen ratios during the first eight days. This close agreement between the nitrate formed in the different treatments was maintained until the concentrations of ammonia became relatively low ; only after the levels of ammonia had been much reduced did the nitrate values begin to diverge, being lowest in the treatment having the highest carbon/nitrogen ratio. Thus although both ammonia and nitrate can be assimilated by the soil organisms, preferential immobilization of ammonia-nitrogen was found in the first half of the experiment.

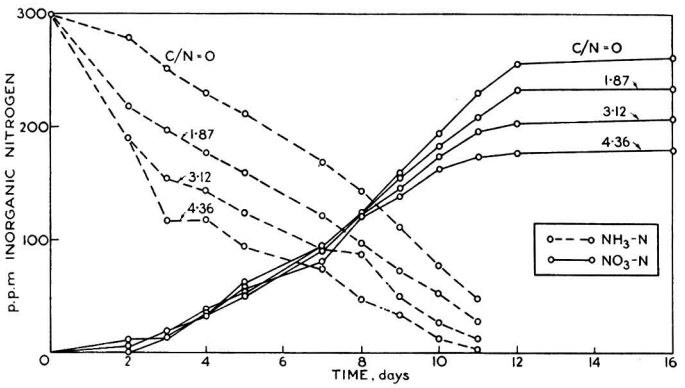


FIG. 1.—*Ammonia- and nitrate-N in a market-garden soil treated with di-ammonium hydrogen phosphate equivalent to 300 p.p.m. nitrogen, and with mixtures of starch and ammonium phosphate having carbon/nitrogen ratios of 1·87, 3·12 and 4·36. The results are shown as differences from the corresponding values for the control soil*

The sums of ammonia and nitrate recovered at different stages of the transformation, expressed as a percentage of the nitrogen added to the soil, are given in Table I for the means of two successive sampling dates. The percentage recovery of nitrogen at each sampling period decreased as the carbon/nitrogen ratio of the added materials was increased. For any one initial carbon/nitrogen ratio the recovery of added nitrogen tended to increase slightly with time, this being most marked at the highest carbon/nitrogen ratio.

**Table I**

*Recovery of added nitrogen in inorganic form in soil treated with di-ammonium hydrogen phosphate and starch at different C/N ratios*

| | % Recovery ($NH_3$-N + $NO_3$-N) | | | |
|---|---|---|---|---|
| Time in days .. .. .. | 3–4 | 5–7 | 8–9 | 10–11 |
| C/N ratio 0 | 80 | 87 | 89 | 92 |
| 1·87 | 70 | 73 | 75 | 79 |
| 3·12 | 59 | 61 | 68 | 68 |
| 4·36 | 47 | 51 | 57 | 58 |

Similar results were obtained in a further experiment with mixtures of starch and ammonium sulphate.

*The effect of mixtures of sucrose and ammonium sulphate on the levels of inorganic nitrogen in the soil*

In subsequent experiments on the immobilization of nitrogen, the pot technique for the incubation of soil samples was replaced by the use of flasks, resulting in increased experimental accuracy. Sucrose was used instead of starch as a readily assimilated carbon compound, and the range of carbon/nitrogen ratios was extended. In one such experiment flasks containing 75 g. of a market-garden soil were treated with ammonium sulphate equivalent to 201·2 p.p.m. of nitrogen, together with sucrose corresponding to carbon/nitrogen ratios of 2·5, 5·0, 7·5 and 10 in the added mixtures. Determinations of ammonia and nitrate were made at intervals on duplicate flasks of soil. The results, expressed as differences from the control soil, have been plotted against the period of incubation up to 10 days in Fig. 2. As shown in Fig. 1, the presence of the added carbon compound caused greater changes in the concentrations of ammonia than of nitrate in the early stages of the experiment. This was particularly the case at carbon/nitrogen ratios up to 5, as used in the preliminary experiments with starch ; at the higher carbon/nitrogen ratios included only in Fig. 2, the nitrate levels at any one sampling date differed more markedly with the carbon/nitrogen ratio of the added materials. The soil organisms thus do not show an exclusive preference for ammonia rather than nitrate at the higher carbon/nitrogen ratios.

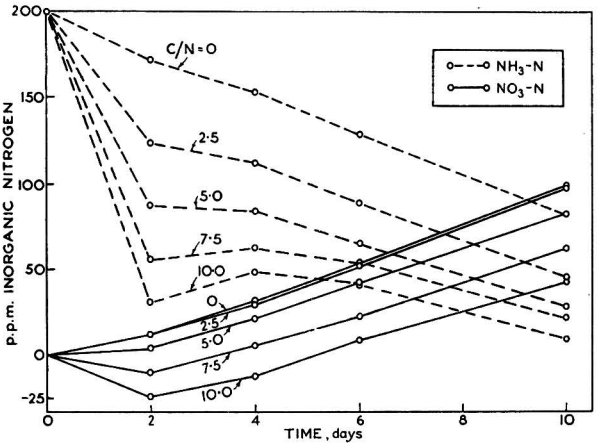


FIG. 2.—*Ammonia- and nitrate-N in a market-garden soil treated with mixtures of ammonium sulphate and sucrose at various carbon/nitrogen ratios. The results are shown as differences from the corresponding values for the control soil*

In the absence of sucrose the concentrations of ammonia decreased steadily as nitrification proceeded. In the presence of sucrose, however, there was a definite break in the curves between two and four days at carbon/nitrogen ratios of 2·5 and 5·0, and even a temporary rise in ammonia levels at carbon/nitrogen ratios of 7·5 and 10, despite the steady conversion of ammonia to nitrate.

The sums of ammonia and nitrate immobilized on incubation with ammonium sulphate and sucrose are given in Table II. The results given for an incubation period of one day were obtained in a separate experiment, using the same soil and treatments as previously described.

## Table II

*Immobilization of inorganic nitrogen ($NH_3$-N + $NO_3$-N) in a market-garden soil treated with ammonium sulphate and different levels of sucrose*

| Time in days .. | 1 | 2 | 4 | 6 | 10 | 24 |
|---|---|---|---|---|---|---|
| Carbon added p.p.m. | p.p.m. $NH_3$-N + $NO_3$-N immobilized | | | | | |
| 503 | 49·6 | 48·5 | 41·9 | 40·9 | 38·4 | 37·5 |
| 1006 | 85·0 | 93·7 | 79·8 | 75·6 | 70·7 | 67·2 |
| 1509 | 122·3 | 139·3 | 116·6 | 105·4 | 97·6 | 91·6 |
| 2012 | 134·6 | 178·8 | 149·5 | 132·7 | 128·4 | 118·8 |

Immobilization of nitrogen in the presence of sucrose is seen to be a rapid process, reaching its maximum in this soil within two days. The break in the curves for ammonia-nitrogen shown in Fig. 2 between the second and fourth days is thus due to the release of some of the immobilized nitrogen after the maximum values found on incubation for two days.

*Immobilization of nitrogen in the form of ammonia and nitrate*

From the data of the preceding experiment, the concentrations of ammonia and nitrate in the soil after incubation for two days have been plotted in Fig. 3 against the carbon/nitrogen ratios of the added mixtures. The level of nitrate-nitrogen in the control soil, receiving neither starch nor ammonium sulphate, is shown on the graph; the concentration of ammonia found in the control soil was extremely low. With mixtures having carbon/nitrogen ratios of 2·5 and 5·0, the nitrogen immobilized was mainly derived from the added ammonium salt, the nitrate concentrations exceeding that of the control soil owing to nitrification. At carbon/nitrogen ratios of 7·5 and 10, however, the nitrogen immobilized was partly derived from nitrate, the concentration

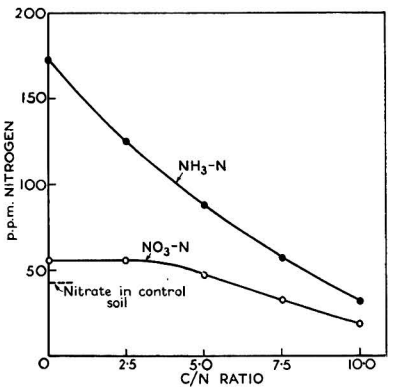


FIG. 3.—*Ammonia- and nitrate-N in a market-garden soil incubated with mixtures of ammonium sulphate and sucrose supplying 201 p.p.m. nitrogen at different C/N ratios. The level of nitrate-N in the control soil is also shown on the graph*

of the latter form of nitrogen being less than in the control soil. As nitrification proceeded on further incubation the concentrations of nitrate exceeded those of ammonia. So long as ammonia remained in the soil at all carbon/nitrogen ratios, however, the curves for ammonia and nitrate retained some similarity in form to those shown in Fig. 3.

Further evidence of the preferential immobilization of ammonia-nitrogen rather than nitrate is presented in Table III. In this experiment samples of two glasshouse soils in moist condition were weighed into flasks in quantities corresponding to 75 g. of oven-dried material. After preliminary analyses, ammonium sulphate and potassium nitrate were added in amounts calculated to raise the concentrations of ammonia and nitrate to 160 p.p.m. of nitrogen in each form. Additional treatments were made with sucrose at six concentrations up to 2000 p.p.m. of carbon added. At the start of the experiment the soil organisms causing the immobilization of nitrogen were thus provided with equal concentrations of ammonia and nitrate, although the balance was somewhat changed subsequently by nitrification during the short period of incubation. The flasks were incubated for two days before analysis, this period having been shown previously to give maximum immobilization under these conditions.

**Table III**

*Ammonia and nitrate in soils after incubation with sucrose for two days. Initial concentrations of ammonia and nitrate-nitrogen raised to 160 p.p.m. in each form*

| Carbon added | Soil 1 | | | Soil 2 | | |
|---|---|---|---|---|---|---|
| p.p.m. | $NH_3$-N | $NO_3$-N | pH | $NH_3$-N | $NO_3$-N | pH |
| 0 | 141·8 | 171·7 | 7·18 | 132·4 | 180·1 | 7·56 |
| 333 | 99·3 | 174·5 | — | 94·5 | 179·7 | 7·51 |
| 667 | 62·5 | 170·9 | 7·07 | 62·5 | 175·5 | 7·49 |
| 1000 | 28·5 | 164·9 | 7·05 | 36·1 | 163·9 | 7·45 |
| 1333 | 11·6 | 148·2 | 7·08 | 11·6 | 154·8 | 7·43 |
| 1666 | 6·4 | 122·5 | 7·11 | 6·3 | 130·5 | 7·56 |
| 2000 | 6·0 | 93·3 | 7·16 | 3·6 | 105·3 | 7·60 |
| Untreated soil | 1·7 | 42·9 | 7·39 | 1·6 | 45·8 | 7·83 |

The results in Table III show that in both soils ammonia was more readily immobilized than was nitrate. Thus the concentrations of ammonia decreased rapidly on incubation with increasing amounts of sucrose, whereas the nitrate levels decreased markedly only at the higher concentrations of sucrose, when the levels of ammonia had already been greatly reduced.

*pH changes accompanying the immobilization of nitrogen in soils*

In the course of experiments on the immobilization of nitrogen in the presence of organic compounds it was noted that appreciable changes in soil pH frequently resulted, particularly when the nitrogen immobilized was derived from either an ammonium salt or nitrate alone. The following experiment was therefore made to compare the effect of immobilization of nitrogen upon the pH of different soils. The soils used for this purpose were: (1) an acid soil, under grass, (2) a glasshouse soil, (3) a market-soil and (4) soil from a shrubbery. Samples of 75 g. of the moist soils were treated with 12 mg. of nitrogen as ammonium sulphate, and incubated with different quantities of sucrose for two days. Further batches of soil were treated similarly except that potassium nitrate was substituted for ammonium sulphate. The pH values of soil suspensions prepared at a 1 : 2 ratio of soil to water are recorded in Table IV.

**Table IV**

*pH of soils incubated for two days with added sucrose and inorganic nitrogen*

| Nitrogen added | Sucrose added mg. C per flask | pH of soils 1 | 2 | 3 | 4 |
|---|---|---|---|---|---|
| 12 mg. of N as ammonium sulphate | 0 | 5·91 | 7·28 | 7·34 | 7·39 |
| | 40 | 5·78 | 7·22 | 7·25 | 7·29 |
| | 80 | 5·68 | 7·23 | 7·17 | 7·25 |
| | 120 | 5·57 | 7·17 | 7·20 | 7·22 |
| 12 mg. of N. as potassium nitrate | 0 | 5·99 | 7·28 | 7·30 | 7·33 |
| | 40 | 6·19 | 7·50 | 7·53 | 7·51 |
| | 80 | 6·53 | 7·74 | 7·62 | 7·78 |
| | 120 | 6·76 | 7·94 | 7·85 | 7·97 |

The results show that immobilization of nitrogen added in the form of an ammonium salt was accompanied by a small but definite fall in pH. Soils (2) to (4), containing carbonate, were relatively well buffered against acidity, but a greater response was found in the somewhat acid soil (1). The increase in pH accompanying the immobilization of nitrate–nitrogen was, in contrast, well marked in all four soils.

In the light of these results some explanation can be given of the relatively small changes in pH previously shown in Table III. The decrease in pH found at the lower levels of added sucrose corresponds to the preferential immobilization of ammonia–nitrogen. As the concentration of ammonia in the soils decreased, however, increasing amounts of nitrogen were immobilized in the form of nitrate, thus again raising the pH of the soils.

*Immobilization of nitrogen in a range of different soils*

The experiments already described show the course of immobilization and mineralization of nitrogen in relatively fertile glasshouse and market-garden soils. Further experiments were accordingly made with a group of eight soils differing widely in origin and having pH values from 8·1 to 3·8.

Samples of each soil were weighed into conical flasks in moist condition, each flask containing the equivalent of 60 g. of oven-dry soil. All samples were treated with ammonium sulphate equivalent to 200 p.p.m. of added nitrogen in the soil, and in addition half the flasks received 2000 p.p.m. of carbon in the form of sucrose. Analyses for ammonia and nitrate were made on duplicate flasks after five periods of incubation ranging from 2 to 18 days. The amounts of nitrogen immobilized are given in Table V.

**Table V**

*Immobilization of nitrogen in eight soils treated with 200 p.p.m. of nitrogen as ammonium sulphate and 2000 p.p.m. of carbon as sucrose*

| Soil | Period of incubation in days 2 | 4 | 6 | 10 | 18 |
|---|---|---|---|---|---|
| | Nitrogen immobilized (p.p.m.) | | | | |
| Glasshouse | 203 | 155 | 143 | 123 | 96 |
| Glasshouse | 205 | 147 | 136 | 119 | 103 |
| Market garden | 177 | 141 | 130 | 120 | 116 |
| Maiden loam | 107 | 92 | 86 | 88 | 72 |
| Shrubbery | 150 | 109 | 120 | 108 | 90 |
| Garden soil | 162 | 147 | 138 | 128 | 116 |
| Rose soil | 187 | 165 | 152 | 146 | 144 |
| Beechwood | 63 | 84 | 89 | 94 | 93 |

The results show marked differences in the ability of the various soils to immobilize nitrogen when compared under standardized conditions of concentration, temperature and moisture content. In seven of the eight soils maximum values for the immobilization of nitrogen were found on incubation for two days, the only exception being the beechwood soil. The latter soil is so acid in reaction, low in essential nutrients and of poor physical structure that rapid biological transformations can hardly be expected.

In Table VI the eight soils have been arranged in the order of their ability to immobilize nitrogen, as indicated by analyses made after incubation for two days.

**Table VI**

*Immobilization of nitrogen in two days, and the pH and phosphate contents of the soils used*

| Soil | Nitrogen immobilized p.p.m. | pH | Phosphate* % $P_2O_5$ |
|---|---|---|---|
| Glasshouse | 205 | 7·57 | 0·39 |
| Glasshouse | 203 | 7·95 | 0·58 |
| Rose soil | 187 | 8·06 | 0·19 |
| Market garden | 177 | 7·55 | 0·14 |
| Garden soil | 162 | 5·26 | 0·04 |
| Shrubbery | 150 | 6·92 | 0·14 |
| Maiden loam | 107 | 5·13 | 0·01 |
| Beechwood | 63 | 3·77 | trace |

* Soluble in 0·5N-acetic acid

The results show a series ranging from the relatively rich glasshouse soils, through the other cultivated but less heavily fertilized soils, down to the maiden and woodland soils. The determinations of pH and available phosphate in the soils, included for comparison in Table VI, show some relationship to the amounts of nitrogen immobilized. Thus relatively alkaline soils of high phosphate content showed the greatest biological activity in this experiment. More extensive experiments on the factors correlated with the immobilization of nitrogen will be described in a subsequent paper.

## Discussion of results

As shown in Fig. 1, the soil organisms concerned in the immobilization of nitrogen show a marked preference for ammonia rather than nitrate when both forms are present in the soil. The preference for ammonia was virtually complete at the lower levels of added carbon compounds, though some nitrate was invariably assimilated at the higher levels of sucrose referred to in Figs. 2 and 3 and in Table III. This marked preference for nitrogen in the form of ammonia is somewhat unexpected since, in the absence of ammonia, nitrate is very readily assimilated by the soil organisms. Heukelekian & Waksman[16] previously noted that certain organisms decomposing cellulose in sterilized soil showed a preference for ammonia rather than nitrate. Shrikhande[17] found that, on incubation of dry cultivated Indian soils with chopped straw (ragi), ammonia was assimilated by the soil organisms in preference to nitrate. A similar result was recently obtained by Boischot & Sylvestre, who showed that in soil treated with straw the utilization of nitrate was inferior to that of ammonia.[18] Nitrogen in the form of ammonium compounds would appear in general to be more suitable as a starting point for the synthesis of cell-constituents. Nitrate may first require reduction before being metabolized,[19] and nitrogen in the form of ammonium salts may thus involve a lower expenditure of energy in its conversion to protein.[20]

In general the immobilization of nitrogen reached its maximum within two days under the conditions of these experiments, after which mineralization of nitrogen predominated. This conclusion is supported by the results for seven out of eight soils in Table V, the only exception being an extremely acid soil having a pH value of 3·8. Further evidence that maximum immobilization occurred on incubation for two days is given in Table II, though the data suggest that the maximum is reached more rapidly at low concentrations of added carbon.

The pH of a soil is decreased by the immobilization of ammonia–nitrogen and increased by immobilization of nitrate–nitrogen. Appreciable changes in the pH of a soil can be produced in this way within two days, the effects due to immobilization of nitrate being particularly marked. Conrad,[21] studying colloid dispersion in the soil, reported a similar increase in the pH of soils treated with nitrate and sucrose. Some analogy can be drawn between these results obtained in the soil and the extensive published data for the growth of various organisms and higher plants

in culture or nutrient solutions. All these studies have in common the basic problem of the passage of solutes through the cell membrane and of their subsequent elaboration within the living cell. Thus the development of acidity accompanying the assimilation of ammonia and the increase in pH when nitrate is assimilated have been reported for fungi,[22], algae,[19] and higher plants.[23] Similar changes in pH were recently recorded by Jansson & Clark[24] for oat-straw decomposing in the presence of inorganic nitrogen. Apart from changes in pH due to the assimilation of nitrogen, the evolution of carbon dioxide from the added organic material will itself tend to reduce the pH of the soil.

Where both ammonia and nitrate are assimilated, the overall change in pH may be relatively small, as shown in Table III ; the pH of the soil was lowered by the immobilization of ammonia, but this trend was subsequently reversed as increasing amounts of nitrate were immobilized at the higher levels of added sucrose.

The rapid changes in pH accompanying the immobilization of nitrogen may be compared with the effects produced more slowly in field soils following the application of nitrogenous fertilizers. Thus ammonium sulphate can cause a considerable increase in acidity, and sodium nitrate some decrease.[25] It is not, of course, suggested that changes in pH resulting from the use of nitrogenous fertilizers are due solely to microbiological transformations, since the processes of ion-exchange play an important part. It may also be noted that the changes in pH accompanying the immobilization of nitrogen are the reverse of those produced by its mineralization. It is well known that the process of nitrification can lower the pH of a soil ;[26] examples of the opposing effect in which the pH is increased by accumulation of ammonia in a partially sterilized soil were encountered in the course of the present investigation.

## Acknowledgments

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# CARBON-NITROGEN RELATIONSHIPS IN SOIL. II.*— Quantitative Relationships between Nitrogen Immobilized and Carbon Added to the Soil

By G. W. WINSOR and A. G. POLLARD

Quantitative relationships between nitrogen immobilized and carbon compounds added to the soil have been studied, using sucrose as the added organic material. A highly significant linear relationship was found between carbon added and nitrogen immobilized. After incubation for two days at 23·5°, this period giving maximum immobilization of nitrogen in the glasshouse and market-garden soils used, one unit of nitrogen was immobilized for every 8·3–10·8 units of carbon added. Part of the added carbon was evolved as carbon dioxide, the ratio of added carbon retained in the soil to nitrogen immobilized being approximately 6 : 1.

## Introduction

In a preliminary paper[1] the nitrogen transformations resulting from the addition to the soil of carbon compounds such as sucrose were discussed. The work now reported is a continuation of these studies, with particular reference to quantitative relationships between carbon added to the soil and nitrogen immobilized.

## Experimental methods

The methods for determination of ammonia and nitrate, available phosphate, pH and moisture equivalent were as previously described.[1] Potash was determined gravimetrically in 0·5N-acetic acid extracts by precipitation as potassium perchlorate. Nitrogen was determined by the Kjeldahl procedure, organic carbon by the method of Walkley & Black[2] and carbonate with the Collins Calcimeter.

The soil perfusion units were assembled as described by Lees.[3]

For the determination of carbon dioxide formation, soil samples (100 g.) were weighed into wide-mouthed conical flasks of 1 litre capacity, the moisture content being raised to 80% of the moisture equivalent. The flasks were closed with rubber stoppers through which passed inlet and outlet tubes fitted with short lengths of rubber tubing and closed with glass rod. A test-tube (6 × 1 in.) containing 10–15 ml. of standardized sodium hydroxide solution was placed in each flask, resting upon the soil. After each period of incubation the flasks were removed from the thermostat chamber and joined into a simple absorption train. A controlled stream of air, freed from carbon dioxide by passage through potassium hydroxide solution, was drawn through the incubation flasks into tubes containing standard alkali. Aeration was continued for 20 minutes, after which the incubation flasks were opened for replacement of the internal absorption tubes. The interchangeable glass tubes containing alkali, as used both in the absorption trains and in the incubation flasks, were titrated with hydrochloric acid after addition of an excess of barium chloride. The titration unit was designed to reduce exposure of the alkaline solutions to the atmosphere, the tip of the burette passing through a rubber stopper which fitted the tubes of alkali. A stream of carbon dioxide-free air, supplied under pressure, gave gentle agitation of the solutions during titration. The standardized solutions of sodium hydroxide used were approximately 0·2N in the aspiration trains and 0·4N for absorption in the incubation flasks. The alkali was titrated with 0·2N-hydrochloric acid, using thymolphthalein as indicator.

All soil samples were incubated at 23·5°. The amounts of nitrogen immobilized in the presence of sucrose have been calculated as the difference in content of inorganic nitrogen between soil samples treated with inorganic nitrogen with and without sucrose. The results are expressed on the basis of oven-dry soil.

Analysis of the glasshouse and market-garden soils used are given in Table I.

* Part I : preceding paper

**Table I**

*Analytical data for market-garden soil A and glasshouse soils B–E*

| | A | B | C | D | E |
|---|---|---|---|---|---|
| Total nitrogen, % | 0·269 | 0·274 | 0·319 | 0·335 | 0·345 |
| Organic carbon, % | 2·56 | 2·50 | 3·01 | 3·01 | 3·34 |
| *Phosphate ($P_2O_5$), % | 0·14 | 0·39 | 0·58 | 0·41 | 0·38 |
| *Potash ($K_2O$), % | 0·05 | 0·07 | 0·05 | 0·08 | 0·08 |
| Carbonate (as $CaCO_3$), % | 0·42 | 0·83 | 3·71 | 1·93 | 1·55 |
| Moisture equivalent | 20·9 | 24·6 | 27·8 | 24·0 | 27·5 |
| pH | 7·6 | 7·5 | 7·8 | 7·5 | 7·6 |

* Soluble in 0·5N-acetic acid

## Results

*The correlation between carbon added and nitrogen immobilized in a market-garden soil*

An experiment was set up in which samples of soil A were treated with ammonium sulphate and with mixtures of ammonium sulphate and sucrose having 16 different carbon/nitrogen ratios from 0·5 to 10. All treatments were applied in triplicate to flasks containing 75 g. of soil, the added nitrogen corresponding to 196 p.p.m. on the basis of oven-dry soil After incubation for 16 days at 23·5°, the flasks were analysed for ammonia and nitrate. The nitrogen immobilized ($x$) has been plotted in Fig. 1 against the amounts of carbon added as sucrose ($y$). The results show a linear relationship, the correlation coefficient $r$ having a value of $+0{\cdot}998$. The regression of $x$ on $y$ is given by the equation $x = 0{\cdot}0602y + 3{\cdot}1$, both carbon and nitrogen being expressed in parts per million on the basis of oven-dry soil. From the regression equation it is calculated that one unit of nitrogen was immobilized for every 16·6 units of carbon added.

The numerical relationship found between carbon and nitrogen in this experiment is not to be regarded as a constant for the soil used, since it varies with the period of incubation. Thus, after immobilization of nitrogen has reached its maximum, the amounts of nitrogen retained in organic form decrease owing to the opposing process of mineralization. This effect may be illustrated by calculations based on an experiment, previously reported,[1] in which soil samples were incubated with ammonium sulphate and sucrose. On plotting the amounts of nitrogen immobilized against the amounts of sucrose added initially the graph is approximately linear for each period of incubation, the slope of the graphs giving values for the amount of carbon required to maintain one unit of nitrogen in organic form. The numerical values so obtained were 11·7, 13·9, 16·2, 16·9 and 18·5 for periods of 2, 4, 6, 10 and 24 days, respectively.

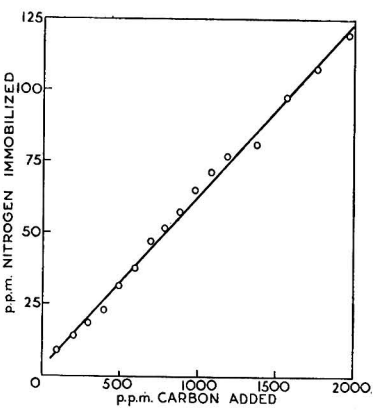


FIG. 1.—*The relationship between carbon added as sucrose to a market-garden soil and the nitrogen immobilized on incubation for 16 days*

*Relationships between carbon added and nitrogen immobilized on incubation for two days*

As discussed in the previous paragraph the amount of carbon which must be added to the soil in order to retain one unit of nitrogen in organic form increases with the period of incubation, the increase being due to mineralization of part of the nitrogen originally immobilized. The relationship between carbon added to the soil and nitrogen immobilized is thus of particular interest when immobilization of nitrogen is at its maximum, this being attained on incubation for approximately two days under the experimental conditions used in this work.

The results of some experiments from which numerical relationships between carbon added and nitrogen immobilized on incubation for two days may be calculated are given in Tables II to IV. Details of these experiments were as follows.

*Glasshouse soils B and C.*—Moist soil was weighed into flasks in quantities corresponding to 75 g. of oven-dry material. After preliminary analyses, ammonium sulphate and potassium nitrate were added in amounts calculated to raise the concentrations of ammonia and nitrate to 160 p.p.m. of nitrogen in each form. At levels of added carbon up to 1000 p.p.m. the nitrogen immobilized was largely derived from ammonia, and the relationship between carbon added and nitrogen immobilized was linear. At higher concentrations of sucrose some departure from linearity was observed, accompanied by the immobilization of increasing amounts of nitrate-nitrogen. The results are given in Table II.

**Table II**

*Immobilization of nitrogen in glasshouse soils B and C on incubation with sucrose for two days*

| Carbon added p.p.m. | Soil B | | Soil C | |
|---|---|---|---|---|
| | $NH_3$-N + $NO_3$-N present | Nitrogen immobilized | $NH_3$-N + $NO_3$ present | Nitrogen immobilized |
| 0 | 313·5 | — | 312·5 | — |
| 333 | 273·8 | 39·7 | 274·2 | 38·3 |
| 667 | 233·4 | 80·1 | 238·0 | 74·5 |
| 1000 | 193·4 | 120·1 | 200·0 | 112·5 |
| 1333 | 159·8 | 153·7 | 166·4 | 146·1 |
| 1666 | 128·9 | 184·6 | 136·8 | 175·7 |
| 2000 | 99·3 | 214·2 | 108·9 | 203·6 |

*Glasshouse soil D.*—This soil was treated with ammonium sulphate in amounts corresponding to 160 p.p.m. of nitrogen in the soil, together with sucrose at six concentrations from 267 to 1600 p.p.m. carbon. The results are given in Table III.

*Glasshouse soil E.*—This soil was treated as already described for soil D, except that potassium nitrate was used in place of ammonium sulphate. The results are given in Table III. The relationship between nitrogen immobilized and carbon added was linear over the whole range of concentrations tested.

**Table III**

*Immobilization of nitrogen in two glasshouse soils incubated with sucrose for two days. Soil D was treated with ammonium sulphate and soil E with potassium nitrate*

| Carbon added p.p.m. | Soil D | | Soil E | |
|---|---|---|---|---|
| | $NH_3$-N + $NO_3$-N present | Nitrogen immobilized | $NH_3$-N + $NO_3$-N present | Nitrogen immobilized |
| 0 | 286·9 | — | 241·9 | — |
| 267 | 259·9 | 27·0 | 219·6 | 22·3 |
| 533 | 230·6 | 56·3 | 193·7 | 48·2 |
| 800 | 202·4 | 84·5 | 166·6 | 75·3 |
| 1066 | 168·8 | 118·1 | 140·0 | 101·9 |
| 1333 | 142·1 | 144·8 | 113·0 | 128·9 |
| 1600 | 120·8 | 166·1 | 86·1 | 155·8 |

*Market-garden soil A.*—This experiment was made to compare immobilization of nitrogen in the same soil after treatment with ammonium sulphate and with potassium nitrate. The nitrogen added in each form was equivalent to 165 p.p.m. on the basis of oven-dried soil, sucrose being added at three concentrations up to 990 p.p.m. carbon. The results are given in Table IV.

From graphs based on the results given in Tables II–IV the amounts of carbon which, when added to the soil and incubated for two days, cause the immobilization of one unit of nitrogen may be calculated. The values so obtained are given in Table V. The results given for glasshouse and market-garden soils show fairly close agreement between the various estimates of the amounts of carbon required in the immobilization of one unit of nitrogen, the values obtained ranging from 8·3 to 10·8, with a mean of 9·5.

*Immobilization of nitrogen, using the soil perfusion technique*

In the experiments already described the soil samples were incubated in flasks, treatments with sucrose and inorganic nitrogen compounds being applied in solutions distributed as uniformly as possible over the surface of the soil. For determination of ammonia and nitrate the

**Table IV**

*Immobilization of nitrogen in market-garden soil A on incubation with sucrose for two days*

Soil treated with ammonium sulphate

| Carbon added p.p.m. | p.p.m. nitrogen present | | | Nitrogen immobilized |
|---|---|---|---|---|
| | $NH_3$-N | $NO_3$-N | $NH_3$-N + $NO_3$-N | |
| 0 | 142·2 | 57·3 | 199·5 | — |
| 330 | 107·1 | 59·4 | 166·5 | 33·0 |
| 660 | 76·9 | 53·8 | 130·7 | 68·8 |
| 990 | 52·8 | 43·2 | 96·0 | 103·5 |

Soil treated with potassium nitrate

| Carbon added p.p.m. | p.p.m. nitrogen present | | | Nitrogen immobilized |
|---|---|---|---|---|
| | $NH_3$-N | $NO_3$-N | $NH_3$-N + $NO_3$-N | |
| 0 | 1·6 | 205·7 | 207·3 | — |
| 330 | 2·0 | 180·3 | 182·3 | 25·0 |
| 660 | 1·8 | 148·5 | 150·1 | 57·2 |
| 990 | 2·0 | 119·1 | 121·1 | 86·2 |

**Table V**

*Amounts of carbon required to immobilize one unit of nitrogen on incubation for two days*

| Soil | Form of inorganic nitrogen added | Carbon added per unit of nitrogen immobilized |
|---|---|---|
| Market garden A | $NH_3$-N | 9·4 |
| Market garden A | $NO_3$-N | 10·8 |
| Glasshouse B | $NH_3$-N + $NO_3$-N | 8·3 |
| Glasshouse C | ,, ,, | 8·9 |
| Glasshouse D | $NH_3$-N | 9·2 |
| Glasshouse E | $NO_3$-N | 10·2 |

entire contents of the flasks were analysed without sub-sampling, thus avoiding possible sampling errors due to uneven distribution of inorganic nitrogen. Owing to the possibility that the distribution of the added materials throughout the soil might conceivably influence the biological activities of the micro-organisms, additional experiments were made using the soil perfusion apparatus described by Lees.[3] Four perfusion units were used, each containing 25 g. of market-garden soil A previously sieved to exclude particles of less than 3 mm. diameter. Three of the units received 100 ml. of solution containing 15 mg. of inorganic nitrogen, half of this being supplied as ammonium sulphate and half as potassium nitrate. Sucrose also was included in the solutions for two of the units, giving carbon/nitrogen ratios in the solutions of 2·5 and 5·0. Distilled water alone was added to the fourth unit, this being run as a control. Perfusion was continued for two days, after which the units were dismantled for analysis. The total inorganic nitrogen in the units is given in Table VI, these values being the sums of ammonia and nitrate in the perfusate and washings, together with that extracted from the soil by leaching with sodium chloride solution.

**Table VI**

*Immobilization of nitrogen in soil perfusion units containing market-garden soil A*

| C/N ratio* | Nitrogen present | | | Nitrogen immobilized mg. |
|---|---|---|---|---|
| | $NH_3$-N mg. | $NO_3$-N mg. | $NH_3$-N + $NO_3$-N mg. | |
| 0 | 6·80 | 7·66 | 14·46 | — |
| 2·5 | 2·66 | 7·45 | 10·11 | 4·35 |
| 5·0 | 0·20 | 6·11 | 6·31 | 8·15 |
| Control | 0·21 | 0·18 | 0·39 | — |

* Initial carbon/nitrogen ratio of solution containing sucrose and inorganic nitrogen

As in the flask experiments previously described, considerably more ammonia than nitrate was immobilized, particularly at the lower carbon/nitrogen ratio. The ratio of carbon added to

nitrogen immobilized in two days was 8·6 at C/N ratio 2·5 and 9·2 at C/N ratio 5. These values are in close agreement with those given in Table V. The main features previously noted in the flask experiments were thus reproduced under the conditions of the perfusion technique.

*Evolution of carbon dioxide in relation to the immobilization of nitrogen*

The experiments already described serve to establish certain numerical relationships between immobilization of nitrogen and the amounts of carbonaceous material added to the soil. Such experiments, however, give little information concerning the utilization of the added carbon by soil organisms. Parallel determinations of the evolution of carbon dioxide and of the accompanying changes in inorganic nitrogen were therefore made.

Market-garden soil A was used in these experiments, 100-g. quantities being weighed into 48 conical flasks of 1 litre capacity. Three different treatments were applied, these being

(1) 10 mg. of nitrogen as ammonium sulphate;
(2) 10 mg. of nitrogen as ammonium sulphate and 50 mg. of carbon as sucrose (C/N = 5);
(3) 10 mg. of nitrogen as ammonium sulphate and 100 mg. of carbon as sucrose (C/N = 10).

The added nitrogen was equivalent to 121·5 p.p.m. and the added carbon to 608 and 1215 p.p.m. at C/N 5 and 10, respectively, on the basis of oven-dry soil. The flasks were incubated for 8 periods of from 1 to 17 days. Determinations of ammonia and nitrate were made in duplicate at each date of sampling, while the evolution of carbon dioxide was measured in quadruplicate with the exception of the last date of sampling. Cumulative totals for the carbon dioxide evolved, expressed as p.p.m. carbon on the basis of oven-dry soil, are given in Fig. 2; corresponding data for the sums of ammonia and nitrate in the soil are recorded in Fig. 3.

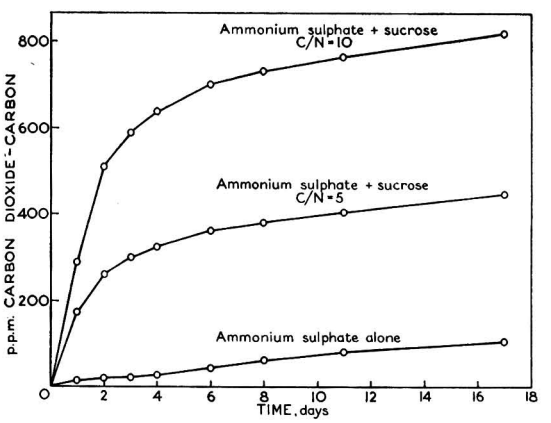


FIG. 2.—*The evolution of carbon dioxide from a market-garden soil treated with ammonium sulphate equivalent to 121·5 p.p.m. nitrogen, together with sucrose to give carbon/nitrogen ratios of 5 and 10 in the added solutions*

The data for the evolution of carbon dioxide show intense microbiological activity in soil treated with sucrose; this activity was particularly marked in the early stages, during which immobilization of nitrogen reached its maximum. The amounts of carbon evolved as carbon dioxide from soil samples treated with sucrose, less that evolved in the absence of sucrose, have been expressed in Table VII as a percentage of the total carbon added at carbon/nitrogen ratios of 5 and 10. Approximately a quarter of the carbon originally added to the soil was converted into carbon dioxide within the first day of incubation, and about 40% within the first two days. As incubation continued, the rates of evolution of carbon dioxide decreased markedly in soil samples treated with sucrose, and after 17 days only 56–58% of the added carbon had been recovered in gaseous form.

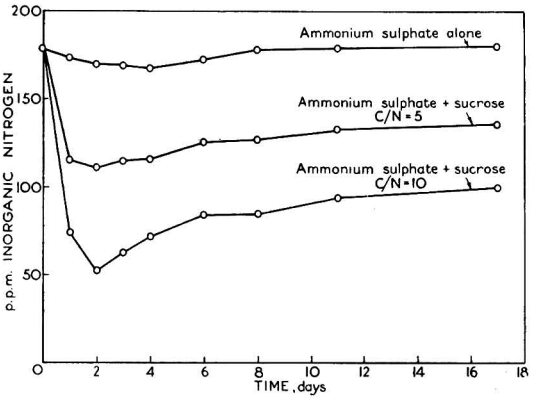


FIG. 3.—*The total inorganic nitrogen present in a market-garden soil treated with ammonium sulphate equivalent to 121·5 p.p.m. nitrogen, together with sucrose to give carbon/nitrogen ratios of 5 and 10 in the added solutions*

## Table VII

*Carbon dioxide evolved from soils treated with sucrose, less that evolved in the absence of sucrose, expressed as a percentage of the total carbon added*

| Time in days | 1 | 2 | 3 | 4 |
|---|---|---|---|---|
| C/N = 5 | 26·2 | 39·7 | 45·6 | 48·7 |
| C/N = 10 | 22·5 | 40·3 | 46·6 | 50·2 |
| Time in days | 6 | 8 | 11 | 17 |
| C/N = 5 | 51·8 | 52·5 | 53·5 | 56·1 |
| C/N = 10 | 53·8 | 54·9 | 56·0 | 58·4 |

By subtracting the carbon evolved as carbon dioxide from the quantities originally added to the soil as sucrose the amount of added carbon still retained in the soil can be estimated, as in Table VIII. The amounts of nitrogen immobilized at the various sampling dates are also included in the Table together with the ratios of carbon retained to nitrogen immobilized. The results suggest that the amount of carbon in the soil associated with the immobilization of one unit of nitrogen is relatively constant, the mean values obtained being 6·0 and 6·2 at C/N ratios 5 and 10, respectively. These values may approximate to the carbon/nitrogen ratio of the micro-organisms concerned together with the organic residues of their decomposition. The actual values obtained are lower than those recorded in Table V, part of the added carbon being evolved as carbon dioxide. Thus in the present experiment the ratio of carbon added to nitrogen immobilized on incubation for two days was 10·3 at both carbon/nitrogen ratios; the ratio of carbon actually retained in the soil to nitrogen immobilized was 6·2, 40% of the added carbon having been evolved as carbon dioxide within the first two days of incubation.

## Table VIII

*Amounts of added carbon retained in the soil after various periods of incubation, the amounts of nitrogen immobilized, and the resultant ratios of carbon to nitrogen*

Soil treated with ammonium sulphate and sucrose at C/N = 10

| Time in days | 2 | 3 | 4 | 6 | 8 | 11 | 17 |
|---|---|---|---|---|---|---|---|
| C retained, p.p.m. | 726 | 649 | 605 | 562 | 548 | 535 | 506 |
| N immobilized, p.p.m. | 118 | 106 | 95 | 88 | 92 | 85 | 80 |
| Ratio of carbon to nitrogen | 6·2 | 6·1 | 6·4 | 6·4 | 6·0 | 6·3 | 6·3 |

Soil treated with ammonium sulphate and sucrose at C/N = 5

| Time in days | 2 | 3 | 4 | 6 | 8 | 11 | 17 |
|---|---|---|---|---|---|---|---|
| C retained, p.p.m. | 366 | 330 | 311 | 292 | 288 | 282 | 267 |
| N immobilized, p.p.m. | 59 | 55 | 51 | 47 | 51 | 46 | 45 |
| Ratio of carbon to nitrogen | 6·2 | 6·0 | 6·1 | 6·2 | 5·6 | 6·1 | 5·9 |

## Discussion

The results shown in Fig. 1 illustrate the very close relationship between carbon and nitrogen transformations in the soil, the correlation coefficient $r$ relating carbon added and nitrogen immobilized being + 0·998.

Immobilization of nitrogen under the conditions of these experiments reached its maximum in approximately two days, after which mineralization predominated. The ratio of carbon added initially to nitrogen immobilized thus increases subsequently with the period of incubation. Particular attention was therefore given to quantitative relationships between carbon added and nitrogen immobilized on incubation for two days, data for five soils being presented in Table V. Quite close agreement was found between these cultivated soils of the glasshouse and market garden, the values ranging from 8·3 to 10·8 units of carbon per unit of nitrogen immobilized. Considerably higher values are, however, found in maiden soils.

The results obtained using soil perfusion technique were 8·6 and 9·2 units of carbon added per unit of nitrogen immobilized in soil A. These values are in close agreement with those given in Table V for soil samples incubated in flasks. Similar values may be calculated from experiments reported by Lees.[4] Thus when soil was percolated with a solution containing sodium nitrate together with sugars, the ratios of carbon added to nitrogen immobilized in four days were 9·4 for sucrose and 9·0 for glucose.

An investigation of the influence of carbon/nitrogen ratio on the immobilization of nitrogen in soil was reported by Osugi *et al.*[5] Soil treated with ammonium sulphate was incubated with varying quantities of glucose, cellulose and rice straw, the amounts of nitrogen immobilized being estimated from analyses at weekly intervals. From the results for glucose at the first date of sampling it may be calculated that approximately 27 units of carbon caused the immobilization of one unit of nitrogen. It is likely, however, that the amounts of nitrogen immobilized on incubation for seven days were considerably less than the maximum values possible, the same authors having previously shown that evolution of carbon dioxide from soil treated with glucose reached its maximum on the second day of incubation.

Further comparisons may be made with the results of investigations of the decomposition of cellulose in the presence of nitrogenous compounds. In the original papers the results are mainly given as units of cellulose decomposed per unit of nitrogen utilized; for purposes of comparison with the present work an arbitrary value of 44·5% carbon in cellulose has been used to relate the quantities of carbon and nitrogen utilized. Thus Hutchinson & Clayton,[6] working with bacterial cultures, found 27·5, 32·3 and 27·7 units of cellulose decomposed for each unit of nitrogen. These values correspond to 12·2, 14·3 and 12·3 units of carbon per unit of nitrogen. Heukelekian & Waksman,[7] working with fungi in sterilized soil, found from 26 to 43 units of cellulose assimilated per unit of nitrogen; these values, increasing with the period of incubation, correspond to 11·6 and 19·1 units of carbon. Jensen[8] reported values of 25–54 units of cellulose (11·1–24·0 as carbon) decomposed per unit of nitrogen by bacteria and fungi in pure culture. The data reported for the decomposition of cellulose in soils and culture media thus correspond to values of from 11 to 24 units of carbon utilized per unit of nitrogen; the lower values are comparable with those obtained with sucrose in cultivated soils in the present investigation.

From parallel determinations of carbon dioxide evolution and nitrogen transformations in soil treated with sucrose the ratio of added carbon retained in the soil to nitrogen immobilized was calculated for various periods of incubation (Table VIII). It was found that the carbon/nitrogen ratios calculated in this way were relatively constant throughout the experiment, and virtually independent of the carbon/nitrogen ratios of the added mixtures of ammonium sulphate and sucrose. Thus for soil treated with a mixture having a C/N ratio of 10, seven carbon/nitrogen values were calculated ranging from 6·0 to 6·4, the mean value being 6·2. Similarly with a mixture having a C/N ratio of 5, the values calculated ranged from 5·6 to 6·2 with a mean value of 6·0. The organic material accumulated in the soil after treatment with sucrose and inorganic nitrogen thus appeared to have a relatively constant carbon/nitrogen ratio.

The addition of readily decomposed organic materials to the soil is known to cause a rapid increase in the numbers of micro-organisms present.[9] The organic material formed in the soil after assimilation of the added sucrose by the soil organisms will in part be present in the

organisms themselves, and partly in the form of organic residues of their decomposition. Data of particular interest in this connexion were obtained by Jensen.[10] The dried cell substance of various organisms, including five soil fungi and an actinomycete, was mixed with sand and inoculated with an extract of garden soil. After incubation for 90 days, during which considerable amounts of nitrogen were nitrified, the carbon/nitrogen ratios of the residual organic materials were determined. The values obtained ranged from 4·3 to 8·4 with a mean value of 6·6. In further experiments the dried microbial substances were replaced by mixtures of sucrose and asparagine. After incubation for 90 days in sand together with added inorganic nutrients the carbon/nitrogen ratios of the organic residues were determined as before. Mixtures of sucrose and asparagine having carbon/nitrogen ratios of 35·6, 22·1 and 8·9 left residues with carbon/nitrogen ratios of 6·6, 6·5 and 5·6, respectively. These values obtained by Jensen, based on actual analyses of the organic residues left in sand cultures, are very similar to the values calculated in the present work for organic material accumulating in the soil (Table VIII). The accumulated organic material had a lower carbon/nitrogen ratio than is normally found for soils themselves. The organic matter of the soil, however, includes not only nitrogenous residues which have undergone transformation by the soil organisms but also nitrogen-free lignin.

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# FUMIGATION OF AGRICULTURAL PRODUCTS. XIII.*— Trials of Onion Seed Treated with Methyl Bromide, and an Improved Method for its Analysis

By O. F. LUBATTI and R. E. BLACKITH

Two varieties of onion seed have been fumigated with methyl bromide over a range of moisture contents. Improvements in the method used for determination of methyl bromide are described. The highest concentration–time product used was applied either as a high dose for a short time or a low dose for a longer period. The germination of seeds in soil is a better measure of the damage done by fumigating moist seed than are laboratory tests on paper pads. Once a seed succeeds in germinating in soil its eventual yield is independent of the treatment received by the seed. Seed dry enough to store with unimpaired germination should not contain more than 10% of moisture and will then withstand concentration–time products of 1100 mg. h./l. irrespective of the way in which this product is made up.

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A slight enhancement of total germination with dry seed properly fumigated was confirmed but is too small to be of economic value. Similarly, the slightly delayed germination of such seed appears to be of little practical consequence. Most of the damage done by fumigating damp seed occurs during the fumigation period, rather than during subsequent storage.

## Introduction

Goodey[1] has shown that 'onion sickness' is caused by the eelworm *Anguillulina dipsaci*. This eelworm is transmitted from infested to clean land on the seeds of infested onions or in dry flower parts accompanying the seed. The parasites can remain dormant for long periods. Goodey also showed that the eelworm could be controlled by fumigation of the seed with methyl bromide.

Lubatti & Smith[2] investigated the action of methyl bromide on onion seed under a variety of conditions, and found that there was a considerable increase of sorption of the fumigant when the moisture content was raised from 10 to 16%. Their germination tests demonstrated that the damage to the seed increased rapidly with increasing moisture content. This damage, which also increased with the dosage employed and with the temperature at which the fumigation was effected, took two forms. In the early stages of the germination test, a pronounced retardation of germination of treated seeds was noted, whilst seed fumigated at the higher moisture contents is killed by the treatment. The minimum sorption of methyl bromide is found when seed is of about 9% moisture content. Although the fragmentary investigations reviewed by Brett[3] suggest that onion seed should be of less than 8% moisture content for prolonged storage, seed in Great Britain is rarely held over from one planting season to another. In consequence, seed containing more than 10% water is often tolerated.

The object of the experiments reported in this paper was to extend these observations to examine the influence of methyl bromide fumigation on the stand and yield of the onions, as well as to measure in detail the delayed germination of treated seeds.

## Experimental

### *Conditioning of seeds*

The two varieties of seed, Bedfordshire Champion and Ailsa Craig, contained originally 10·3% and 9·4% moisture respectively. In order to obtain a range of seed of different moisture content, 100-g. batches of the two varieties were allowed to take up moisture over aqueous solutions of potassium hydroxide in a metal cabinet, or dried in a desiccator over calcium chloride.

The conditioning cabinets are those described by Brown.[4] There are four trays in each cabinet capable of containing 50 g. of seed each, viz. 100 g. of each variety. By means of a timing device the air in the cabinet can be stirred for a period of 5 minutes every 55 minutes, in order to secure adequate mixing without noticeable heating.

The process was carried out in a constant-temperature room at 20°. The progress of water absorption was determined by observing the increase in weight of about 5 g. of seed contained in a small muslin bag which could be withdrawn from the box through an opening and weighed in a weighing bottle.

In the case of the dry batches the loss of weight was ascertained at intervals by withdrawing from the desiccator the 100 g. of seeds contained in a muslin bag and weighing them enclosed in a light aluminium box.

After conditioning, the different batches of seed were stored in glass stoppered jars and the moisture content was then accurately determined.

### *Moisture determination*

The method employed was basically similar to the two-stage air-oven method specified in the Handbook of the Official Grain Standards of the U.S. Department of Agriculture.[5] In the first stage, approximately 20 g. of seeds were taken in duplicate, and from each of these duplicate portions, approximately 5 g. of ground seed were obtained for the final stage. In the first stage, metal boxes 10 cm. diameter, 2·5 cm. deep were used; for the second stage aluminium boxes 6 cm. diameter, 1·5 cm. deep.

The moisture content of the seeds used is given in Table I. This range straddles the critical levels for which sorption of methyl bromide on onion seed is minimal.[2]

*Fumigation of the seeds*

The glass jars containing the seeds were stored in a constant-temperature room at 20°. One portion of 2 g. was removed from the jar by means of a calibrated metal spoon and each portion was introduced as rapidly as possible into a small muslin bag. Two muslin bags, one for each variety, in equilibrium with the same relative humidity, were then placed in the appropriate fumigation chamber.

This type of chamber, ' Turtle Chamber ', has been described by Lubatti.[6] Chambers of two capacities were employed in these experiments, viz. approximately 1000 c.c. and 1700 c.c. Into each chamber, together with the seed, was placed an ampoule of methyl bromide, containing a known weight of the fumigant. These ampoules were prepared as described by Lubatti.[7] The larger chambers were used for the longer periods of exposure to avoid the use of ampoules containing an unduly small amount of bromide.

The chamber was stoppered and left in the constant-temperature room for 48 hours. It was assumed that over this period of time the seed under test would condition the atmosphere of the chamber. The ampoule, previously weighted with a piece of glass rod, was then fractured in the smaller section of the chamber.[6] The chamber was inverted ten times and allowed to stand in the constant-temperature room for the allotted time. Duplicate gas samples were collected from the chamber about 15 minutes before the end of fumigation.

The samples were withdrawn from the chamber through a glass capillary tube, 1 metre long, attached to one of the taps by a short piece of rubber tubing and a butt joint. The method of gas sampling was that developed by Page.[8] The sampling flask, about 50 c.c. in capacity, was of the type described by Wade.[9]

*Determination of methyl bromide*

*Catalytic combustion apparatus.*—The methyl bromide contained in the sampling flasks was determined by the catalytic combustion method employed by Lubatti & Harrison.[10] A modification of the original combustion unit is shown in Fig. 1. The body of the new former (1) consists of a silica tube 12 mm. diameter, and 6 mm. bore. In the grooved portion, 170 mm. long, there are two parallel spiral grooves about 1 mm. deep terminating near two small silica buttons (2). The plain portion ends, at the bottom, in a smaller tube 35 mm. long, 7 mm. diameter, 3 mm. bore. The former is cemented into a block of laminated plastic material (3). This is 55 mm. long and 32 mm. diameter at the lower end ; at the top, over a length of 30 mm., this block is turned down to 22 mm. diameter ; the intermediate portion, 12 mm. long, is 28 mm. diameter. The block carries two terminals (4) connected by an internal copper wire (5) of 20 s.w.g. to two platinum leads (6) of 30 s.w.g. These two leads, in the proximity of the fastening silica buttons (2) fused on the former, are attached to the loose ends of the catalytic platinum wire (39 s.w.g.) which is housed in the bottom of the spiral groovings. This wire, about 2·5 m. long, is continuous, rising from one of the buttons (2) to the top of the former and then descending to the other button. Fitting snugly over the grooved portion of the former is the outer silica jacket (7). This jacket opens out 1 or 2 mm. above the fastening buttons (2) to a diameter of 22 mm. so as to fit closely over the upper part of the laminated plastic block (3). In the region where it narrows down, the enlarged portion of the jacket carries a side tube (8) which is bent downwards to run parallel to the former (1). This tube (8) is of the same diameter and bore as the narrow tube of the former, viz. 7 mm. diameter and 2 mm. bore, and ends in a B10 socket. The silica jacket (7) is connected by a rubber sleeve (9) to the intermediate portion of the plastic block (3). The joint is made airtight by painting over with shellac varnish. The narrow portion of the silica jacket (7) is covered with lagging consisting of a tube (10) made of asbestos paper.

The catalytic combustion tube is one of the units of a system (Fig. 2) consisting of an air purifying train (activated charcoal) A, a flowmeter B, a trap C, the sampling tube D and a by-pass E, a three-way tap F, a mixing chamber G, the catalytic combustion tube H, a bubbler I, a regulating valve J, and a suction unit K, provided with a leak valve L.

The bubbler Fig. 3 (I) consists of a glass tube A 100 mm. long, 22 mm. outside diameter. The central tube B, provided with a B10 cone, is connected to a small sintered glass filter C (10 mm. diameter; porosity 1) which is set at an angle of 45° at the bottom of the tube. At the shoulder there is a side-arm D provided with a bulb; the diameter of this tube is the same as that connected to the sintered glass filter, viz. about 6 mm. outside diameter, and of 4 mm. bore.

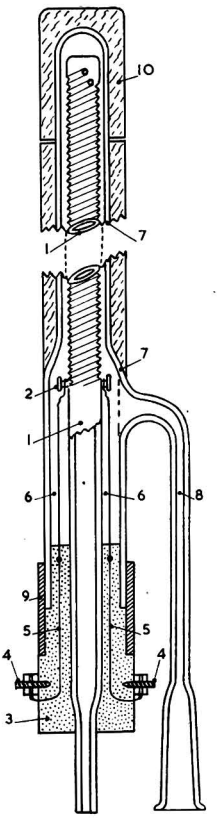


FIG. 1.—*Catalytic combustion apparatus*

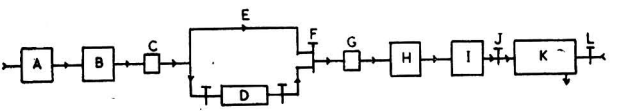


FIG. 2.—*Gas analysis train*

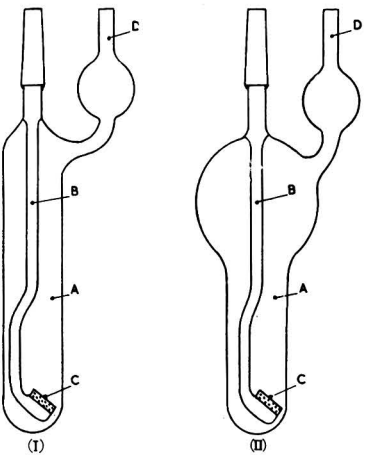


FIG. 3.—*Bubblers from catalytic combustion unit*

*Method of operation.*—The terminals of the combustion tube are connected to the mains supply, in series with a sliding resistance. In our particular apparatus a pair of catalytic tubes in parallel is connected to a sliding resistance in series. With a 240-v supply the resistance employed is 85 ohms at 4 amp.

The sliding contact is adjusted so as to bring the temperature of the platinum filament to about 700°; this temperature does not need to be exceeded as has been shown by Call.[11] The glowing filament can be observed by lifting the upper portion of the asbestos lagging (10) which forms a cap. The tip of the jacket (7) is made of transparent silica to facilitate inspection. About 20 minutes are required to reach this temperature.

The sampling tube is connected by means of two short pieces of rubber tubing and butt joints in the position D (Fig. 2). Five c.c. of 0·1N-sodium hydroxide are introduced into the bubbler through the side-arm by means of a syringe provided with a thin plastic tube. The two members of the B10 joint are used to connect the furnace H and the bubbler I. The side-arm of the bubbler is connected by rubber tubing to the regulating tap J and the suction unit K.

Tap F is turned to the by-pass. The suction is regulated so as to draw air through the system at the rate of about 100 c.c. per minute. In our arrangement the suction system is attached to two combustion trains. At first the leak valve L is closed in getting the required rate of flow approximately, the final adjustment being made by the regulating tap J carried by each train.

When the flow is steady, the tap of the sampling tube nearer to the catalytic tube is first opened, then the other tap, and finally air is swept through the sampling tube, the tap F having been turned in the appropriate direction. The air of the sampling tube, laden with methyl bromide vapour, is carried to the mixing chamber G, then to the central tube of the combustion unit H. The methyl bromide rises with the air in the internal cavity of the heated former. Its combustion is completed when, escaping from the top of the former, it makes contact with the heated platinum spiral situated in the air space between the jacket and the former. The bromine, resulting from the combustion of the methyl bromide, issues from the tube at the side of the jacket and is subsequently absorbed by the sodium hydroxide solution in the bubbler.

The flow through the system is maintained at the rate of 100 c.c. per minute for 5 minutes. During this time the bulk of the bromine resulting from the combustion is carried over. The process is continued for another 10 minutes at the rate of 250 c.c. per minute.

One bubbler has been found sufficient when the aeration of the sampling tube (100 c.c. or less) is carried out as indicated. Experiments carried out over a range of concentration varying between 30 and 200 mg. per litre with two bubblers in series have shown that the amount of bromine collected in the second bubbler is negligible.

When chambers of 1000 and 2000 c.c. capacity containing fumigated materials are being aired, higher rates of flow are necessary. In this case two catalytic tubes and two bubblers, alternating, are employed[6] in series. The bubblers designed by Wade[9] for this purpose are provided with an expansion, Fig. 3 (II), which prevents frothing over. Rates of flow up to 2 litres per minute are then permissible.

*Electrometric titration.*—The rate of flow through the train is slowed down and the bubbler disconnected. The liquid contained in the bubbler is then transferred to a small beaker or cup (40 mm. diameter, 55 mm. high, approximately). A length of rubber tubing is attached to tube B (Fig. 3) leading to the sintered glass disc, and the liquid is blown gently into the cup while holding the tip of the arm D against its side. Washing is carried out by connecting the rubber tubing to a washing-bottle spout, and transferring from it 4–5 c.c. of water. The liquid collected in the bubbler is then again transferred to the cup by blowing directly through the rubber tubing. Washing is repeated three times.

The liquid in the cup is evaporated on the water bath, the residue is suspended in 5 c.c. of water, 1 drop of neutral red is added and the liquid is acidified with N-sulphuric acid. The liquid is then titrated potentiometrically as indicated by Wade.[12]

The measurement of the potential at the equivalence point is carried out with a suitable titrimeter. The battery-operated meters of Russell[13] and of Anderson & Hindman[14] as well as the mains-operated meter of Scroggie[15] have been satisfactorily employed.

Call, working in this laboratory, has developed an inexpensive titrimeter, the circuit of which is shown in Fig. 4. When used with a stabilized power supply such as that described by Lampkin,[16] the zero drift is less than 0·5 microamp. for 10% change in main voltage. However, the meter can be used without such a stabilized supply, when the zero drift will be slightly greater.

*Determination of the concentration–time product*

In the glass fumigation chambers used in these experiments there is a slight fall of concentration due to sorption of methyl bromide on the seed and the adsorption on the walls of the chamber. The fall was of the order of 4% for the higher concentrations (200 mg. per litre) over a period of six hours; there is an appreciably smaller fall for the lower concentrations.

In these experiments, the mean of the original nominal concentration calculated from the weight of methyl bromide added and the concentration obtained from the duplicate determination at the end of fumigation was assumed to represent the average concentration prevailing over the period of fumigation.

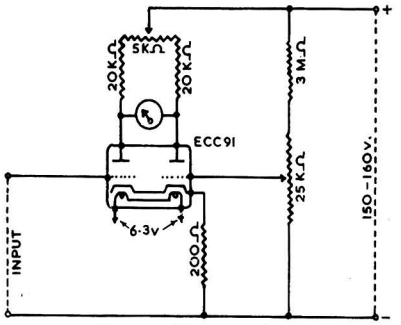


FIG. 4.—*Titrimeter circuit*

The fixed resistors 200 Ω, 20 kΩ, are at high stability. Actual values are not critical. 5 kΩ resistor is a wire-wound preset potentiometer. 25 kΩ resistor is a wire-wound potentiometer. Grid leads are carried under chassis. The meter is provided with a suitable shunt and switch.

The concentration–time products obtained, as calculated from the average concentration thus obtained, are given in Table I.

Two of the treatments gave rise to the same concentration × time product (1100 mg. h./l.) but were made up of a short exposure period and high concentration, and conversely. The form in which a given concentration × time product is presented to seeds has been shown to influence the amount of fumigant sorbed.[17]

The discrepancies among the concentration–time products are due to the difficulty of preparing ampoules containing exactly the required amount of fumigant. These discrepancies are not serious enough to interfere with the statistical computation.

## Table I

*Moisture content and fumigation treatment of conditioned onion seed*

| Moisture content | | Duration of treatment (h.) | Concentration–time product attained (mg. h./l.) |
|---|---|---|---|
| Bedfordshire Champion % | Ailsa Craig % | | |
| 8·1 | 8·1 | 24 | 814 |
| | | 6 | 1157 |
| | | 48 | 1139 |
| 10·6 | 10·3 | 24 | 817 |
| | | 6 | 1164 |
| | | 48 | 1111 |
| 12·6 | 11·8 | 24 | 824 |
| | | 6 | 1191 |
| | | 48 | 1164 |
| 17·2 | 16·2 | 24 | 814 |
| | | 6 | 1149 |
| | | 48 | 1073 |

*Aeration and storing of fumigated seed*

At the end of the appropriate period of fumigation, the two muslin bags removed from the fumigation chambers were allowed to air for a period of about one hour. The content of each bag was divided into two parts, each of 1 g.; one part was transferred to a small glass tube and closed with a paraffined bark stopper, whilst the other was allowed to stand in a similar tube which was left uncorked for 48 hours in the atmosphere of the laboratory. The period between fumigation and planting was not quite the same for all batches—it varied between 30 and 40 days. So far as the seeds planted directly in the open are concerned, a further five-week period during which virtually no rain fell might well be added to the storage period.

Although the overriding importance of the moisture content of the seed to be fumigated is now well known, practically nothing is known of the relative importance of this moisture at the time of fumigation or during the subsequent storage period. To this end, as just described, half of each batch of seed was dried to the normal moisture content (about 8%) after fumigation, the remainder being kept at the moisture content at which it was fumigated until it was sown. The first response of the seeds to be measured was the delayed germination, which was estimated by the standard germination test previously described by Lubatti & Smith.[2] This response was measured by the product of the deficiency of the number of treated seeds germinating, relative to the controls of the same variety and moisture content, and the number of days for which the

deficiency persisted. The germination tests were continued until all the seeds could be classified as germinated or dead. The deficiency in the various treated batches was then corrected for the number of dead seeds, which was recorded as the second response. Germination was estimated after one month had elapsed after the fumigation. Batches of 100 seeds were used for each of the 64 categories of seed specified by the experimental design.

The seeds set aside for the field trials were divided into two equal lots, at the time of the first germination test (April, 1954). One lot was planted directly in the open air, in light loam. The other was planted in seed boxes and kept in a cold glasshouse until early June, when the seedlings were transplanted to the field under the same conditions as those sown directly in the open. The lay-out is described in ' Design of field experiments ' below. The plots were weeded manually.

The summer of 1954 was so wet and cold that onion growing was unusually difficult. Our original intention was to estimate the yield of those onions suitable for storage, but almost all the crop was ' bull-necked '. For this reason, the crop was lifted green, in September, since the persistent rains made the normal practice of bending over and drying the plants in the field impracticable. The plants were laid on hessian and allowed to dry for two weeks under cover. Preliminary trials showed that it is most important to rub out all the soil from between the bulb roots, as traces of this adherent can contribute more to the apparent weight than does the voluminous ' top ' to a bull-necked onion. Such imperfectly matured onions require more careful cleaning than do properly ripened specimens, as the root system is usually still luxuriant.

A further difficulty was the poor quality of the Ailsa Craig seed, which averaged 58·8% germination when tested in April, although, no doubt, above the minimum saleable figure of 60% when received. This batch of seed was used by several market gardeners and others in the district surrounding the Field Station, and the crop was in several instances abandoned as an economic failure. The germination in soil, averaged over all control plots sown with Ailsa Craig seed in our experiments, was only 10%. When this state of affairs was revealed by the initial germination tests, the size of the field trials was doubled in compensation.

*Design of the field experiments*

The complicated design adopted for the field trials was made necessary because a number of the factors investigated were of interest only to the extent that they might modify the effects of the fumigation treatments. For instance, the differential yield of the two varieties in this trial, or of the directly sown and transplanted material, is an agronomic matter outside the sphere of interest of this investigation. But these comparisons were included in order to broaden the inductive basis of this experimental investigation of the effects of the methyl bromide fumigation. Moreover, the difference in the ways in which the highest concentration–time product was made up seemed likely to need particularly precise estimation of the responses of the seeds as it was unlikely to be commensurate with the differential responses to the doses used.

The field trial was, therefore, conducted at four levels of accuracy. The basic design was the $4 \times 3 \times 2$ factorial lay-out, accommodating the four moisture contents, the three main levels of concentration $\times$ time product, and the two conditions of storage prior to sowing and after fumigation. There were six complete replicates of this basic lay-out, providing six blocks of 24 plots each. Further, each plot containing onions receiving the highest concentration $\times$ time product was split in two, each half accommodating onions treated in one of the two ways which provided this highest dosage. Three complete replicates were devoted to the glasshouse sown plants and three to those planted directly. This comparison is referable to the variability between blocks. Finally, each block of 32 plots (including split-plots) was divided into two sub-blocks of 16 plots each, so that the comparisons confounded between the sub-blocks were complicated high-order interactions which may be recovered if so desired by the method of analysis given by Li.[18] In these experiments the comparison between the two varieties was confounded with the sacrificed interactions, one half of each block being sown with one variety, the remainder with the other variety. Each plot contained 30 plants, at 6-in. intervals, the same distance lying between plots, so that a sub-block occupied only 15 ft. $\times$ 8 ft. The whole-plot error variance was three times that for adjacent half-plots, and was only marginally less than the sub-block error variance.

*Germination tests: standard laboratory technique*

The results of the germination tests, by the National Seed Testing Station method,[2] fall into two categories—the estimates of mortality and of delayed germination. Because of this delay, the tests had to be prolonged much beyond the 12 days of the official method; indeed, seeds were still germinating 39 days after the test was begun.

Somade[19] has observed that methyl bromide fumigation prevents fungal attack on groundnuts germinated in sand. No such inhibition was noticed in the experiments with onion seed. Although dead seeds quickly give rise to copious growth of mycelium, not all seeds covered with fungus were dead. The only safe criterion of death is autolysis, which process ruptures the seed-coat and liberates the usually yellow-green liquid contents. Such autolysed seeds must be promptly removed from the Petri dishes containing, initially, 100 seeds, if the test is to be prolonged for a month or more, otherwise fungal growth renders examination impracticable.

*Germination tests: emergence from soil*

Unless the laboratory tests are truncated, no effective distinction can be made between germination capacity and germination energy. These qualities may loosely be described as viability under ideal conditions and under natural conditions respectively. When, as here, seeds are given chemical treatments which may impair germination, it is of obvious importance to be able to distinguish those seeds so weakened as to be viable only under the special laboratory conditions. At the same time outdoor field trials may, as in 1954, provide unrepresentative severe conditions for germination tests. As a compromise, three replicates of the field trial were sown in seed-boxes in a cool glasshouse. Each box contained 30 seeds, set on potting soil and covered lightly with sifted soil, one plot of the field trial corresponding to each box.

The criteria of germination are necessarily different: in the laboratory tests the appearance of the root is recorded from inspection of the bare seed, whereas the appearance of the green shoot is recorded in the experiments in soil.

In these glasshouse tests the germination records were made three weeks after sowing. The remaining three replicates were sown directly into the soil where they were to grow. For these replicates germination was not complete until five weeks after sowing because of a period of dry, cold weather. By so providing conditions of widely different severity, the inductive basis of these experiments was broadened.

The germination counts, made as described above, are to be distinguished from the number of plants weighed at the end of the experiment. As in most field experiments, a few losses were suffered from causes having nothing to do with the treatments being examined. The analysis of the germination counts served to provide the basis of the interpretation of the influence of fumigation on germination. However, in order to examine the comparable influence of fumigation on yield, we need to separate the component of yield differences attributable to germination differences. For the analysis of covariance which secures this separation, the ultimate stand, rather than the initial germination, has to be employed, taking care that a form of analysis is used which accommodates without bias the influence of the treatments on germination.

## Results

*Laboratory germination tests*

The general pattern of germination of fumigated Ailsa Craig seed is shown in Fig. 5 and for fumigated Bedfordshire Champion seed in Fig. 6. Throughout this paper only those conclusions are given which can be substantiated by an analysis of variance of the data, followed where appropriate by an analysis of ranked mean values according to the scheme of Tukey.[20] Probability levels are attached to each conclusion.

The overriding importance of the moisture content of the seeds makes itself felt in four ways. The most important of these is the rapid deterioration of unfumigated seed. Seed of 18% moisture content died in storage when of variety Ailsa Craig and was of greatly impaired germination in the case of Bedfordshire Champion. Even seeds of 12% moisture content showed a significant reduction of viability for the variety Ailsa Craig ($P < 0.05$) though not for Bedfordshire Champion ($P > 0.05$). Seeds of 8% and 10% moisture content, of either variety, did not differ significantly in germination.

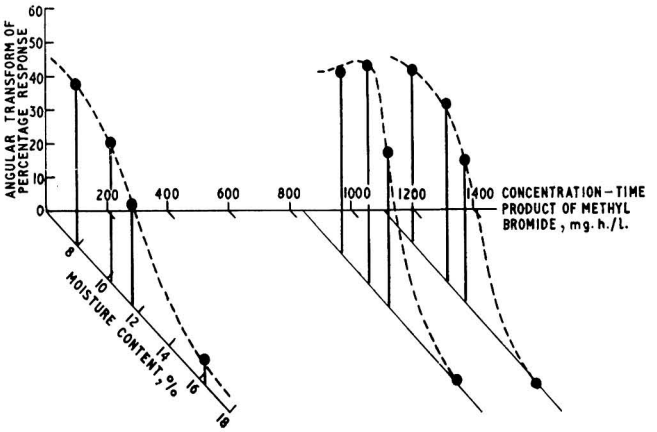


FIG. 5.—*Germination of fumigated Ailsa Craig seeds*

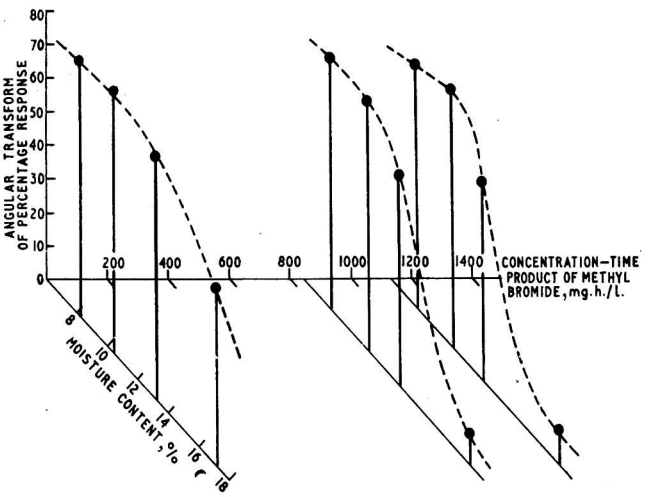


FIG. 6.—*Germination of fumigated Bedfordshire Champion seeds*

The action of the fumigant is greatly influenced by moisture in the seed. Somade[19] found that groundnuts were stimulated by low doses of methyl bromide to give enhanced germination, whereas higher doses tended to kill the seed. The stimulant action of methyl bromide on onion seed can also be demonstrated. At the lower rate of dosage (840 mg. h./l.), the percentage germination of the two drier categories of Ailsa Craig seed is enhanced by 6·5% for seed of 8% moisture content and 7·5% for seed of 10% moisture content. The combined evidence of these groups demonstrates stimulation ($P < 0{\cdot}05$). At 12% moisture content, the germination is virtually independent of dosage, but at 18% even the lower dose produced a dramatic fall. Despite the much lower initial viability of the Ailsa Craig seeds, compared with Bedfordshire Champion seed, the two varieties react alike to the moisture content, dosage of fumigant, and according to whether they are dried out after fumigation or not ($P > 0{\cdot}05$ in each instance).

Drying some of the seed immediately after fumigation enables the distinction to be made between damage during fumigation and damage during storage. The overall effect of post-

fumigation drying is slight ($P < 0.05$) but significant. As we should expect, the improved germination of such dried seeds is greatest when these are initially of the highest moisture content ($P < 0.01$). A feature worthy of fuller discussion later is the fact that post-fumigation drying loses some of its effectiveness with increasing dosage of fumigant ($P < 0.05$).

No distinction could be made between the two ways of making up the highest concentration–time product, both of which gave equivalent results throughout this part of the experiment.

*Delayed germination of fumigated seed*

The statistical significance of delayed germination in treated batches of seed was assessed by estimating the variability of unfumigated dry seed in this respect, regarding as significant of real delay only those accumulated deficiencies of seed-days which exceeded twice the standard error of unfumigated batches of seed. This form of analysis avoids the problem raised by the high serial correlation of germination counts, to which sufficient attention is rarely paid. Significant delays may be compounded of a few viable seeds with abnormally long latent periods, or by the bulk of a batch of seeds exhibiting more moderately extended latent periods.

Of the Ailsa Craig seeds, delayed germination was found principally with the driest batches when these received the highest concentration–time products of fumigation treatment. Fumigated Bedfordshire Champion seeds, on the contrary, were most delayed in their germination when of the highest moisture content. Although the germination of all fumigated batches of this variety was delayed to some extent, the delay was significant only for seed of the two highest moisture contents chosen. Neither variety responded differently when dried immediately after fumigation or when allowed to remain at the moisture content at which it was fumigated. The germination of unfumigated seeds, however moist, was not delayed, although they were on the point of death.

*Field germination tests*

The conclusions drawn from seeds sown in soil substantially confirm those from seeds set on paper pads in the laboratory. The soil experiments are even more clearly defined because weakly seeds failed to germinate under the more rigorous conditions, and the treatment differences take on something of an all-or-nothing quality.

Of the seeds fumigated at 18% moisture content, none germinated of either variety, nor did any unfumigated seeds of this moisture content save for a few Bedfordshire Champion seeds and those of either variety which had been dried after wetting. Enhanced germination relative to the controls was again found when lightly fumigated dry seeds were grown in soil as in Petri dishes ($P < 0.05$).

The importance of drying seed after fumigation was diminished in the field trials, only the seeds of the highest moisture content showing any appreciable benefit from dry storage ($P < 0.05$).

The differing cultural conditions brought out varietal differences not shown in the laboratory tests. Seeds sown directly in the open suffered more from the effects of a high moisture content than did those sown in the glasshouse, and seeds of Ailsa Craig variety more so than those of Bedfordshire Champion variety ($P < 0.01$ and $P < 0.001$, respectively). Curiously, this varietal distinction is reduced, rather than enhanced, by the fumigation treatments ($P < 0.01$) but in view of the complex results possible when a treatment may either stimulate or depress germination, according to the levels of both fumigation dosage and moisture content, prediction of results is an unprofitable process. Moreover, the result obtained accords with the fact that the more stringent conditions in the open air did not enhance the effect of fumigation ($P < 0.05$).

Although seeds of Ailsa Craig variety germinated less well than those of Bedfordshire Champion variety in the glasshouse, and both varieties did better in the glasshouse than in the open air, there is no clear evidence that the reduction of the germination of Ailsa Craig seeds in the open was proportionately the greater, this interaction not quite reaching the 5% significance level.

*Yields of the crop*

The preliminary analysis of variance of the yields of the crop disclosed many of the differences already noted from the germination trials, but, when the yields were examined by means of

an analysis of covariance, all such differences were found to be attributable to the influence of the treatments on the number of plants surviving rather than on the yield per surviving plant.

## Discussion

The result of immediate practical importance is that so long as a seed manages to germinate in soil, it will produce an onion of which the weight is independent of the treatment received by the seed. Further, differences in the severity of the cultural conditions are of less importance in assessing fumigation damage than is the distinction between laboratory tests on filter paper pads and tests in soil. Seeds whose germination energy has been impaired may yet retain the capacity to germinate in the laboratory, although for practical purposes they are unlikely to survive in the field.

The delayed germination of fumigated seeds seems to be of little economic importance, although a weather sequence is conceivable in which unfumigated seed might just succeed in establishing itself before a drought afflicted the fumigated seed. The evidence from these onion seed tests, together with the groundnut experiments of Somade,[19] suggest that light doses of methyl bromide enhance germination. The economic value of this enhancement, whose mechanism is at present obscure, is slight.

The recommendation of a safe concentration–time product of methyl bromide follows the lines already noted by Somade for groundnuts. The rapid deterioration of seed, unless it be dry, enables one to make the recommendation that seed suitable for storage (i.e. of less than 8% moisture content) will tolerate a concentration–time product of 1150 mg. hr./l. comfortably. Moreover, considerable latitude is available in the relative contributions of time and of concentration in making up the concentration–time product.

The distinction between the damage done by storing seed while moist and that done by fumigating it in the same state was well brought out by the device of drying part of the seed after fumigating. The deterioration of moist seed was arrested in the dried samples, but the harmful effect of methyl bromide fumigation on such seed was but slightly mitigated. We may conclude that most of the damage done by methyl bromide takes place during the actual fumigation, with a small and economically negligible component of damage from reactions continuing after the seed has been aired.

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# STUDIES OF SPRAY DEPOSITS. I.—Effect of Spray Supplements on the Tenacity of a Copper Fungicide

By E. SOMERS

The influence of 47 materials on the tenacity of cupric oxide on a cellulose acetate surface has been determined. From these screening tests, seven of the most promising stickers, agar, linseed oil, lime casein, polyvinyl acetate (PVA), coumarone resin, rubber latex and polyvinyl chloride (PVC) were used as supplements to cupric oxide in a potato blight field trial. Although most of the supplements increased the tenacity of the cupric oxide, little, if any, improved blight control was observed. Linseed oil and rubber latex were also used as supplements in a field trial on tomato foliage and the copper residue level was determined chemically.

The effect of varying the supplement concentration of boiled linseed oil, PVA and rubber latex on the tenacity of cupric oxide on cellulose acetate and bean leaves was determined. The most effective concentrations were the same for both surfaces. Sedimentation analysis showed that both boiled linseed oil and rubber latex flocculated the cupric oxide suspension, whilst PVA at a high concentration increased its dispersibility.

Assays *in vitro* against *Alternaria tenuis* showed that, of the seven CuO-supplement treatments listed above, PVC and PVA were the only supplements that did not decrease the fungitoxicity of the cupric oxide. The physico-chemical aspects of this inhibition are discussed. It is suggested that PVA, at sufficiently high concentration, should prove to be a suitable sticker for copper fungicides.

## Introduction

The value of protective fungicides applied to foliage is largely determined by their ability to resist the weathering action of rain, sun and wind. In recent years there has been a tendency to replace Bordeaux mixture by copper compounds such as the oxide, basic carbonate, and oxychloride and various supplementary materials, termed ' stickers ', have been added to the spray suspensions to increase the adhesion of the fungicide to the leaf surface. The resultant effect is assessed in terms of the tenacity of the fungicide, which is defined[1] as the ratio of the weight of fungicide residue at a given time to its initial deposit.

Fajans & Martin[1] investigated the effect of a series of chemicals on the tenacity of cuprous oxide and iodide on artificial surfaces. They found that both oils and those substances which gave insoluble residues improved the tenacity, whilst surface-active agents exerted a deleterious effect. A field experiment showed that supplements which increased the tenacity and uniformity of a cuprous oxide deposit also increased its control of potato blight. Harry[2] summarized the literature on the use of stickers with pesticide sprays and concluded from this review and from his own work that few of the materials which increase the tenacity of fungicides in laboratory and field trials improve their control of disease. Garman[3] found that bentonite–skim milk, soya-bean oil, and some synthetic resin emulsions increased the tenacity of lead arsenate–sulphur sprays on privet and apple leaves, and that in field trials with the first two materials improved biological control was obtained. Physical methods were used by Green[4] to determine the tenacity, on glass slides, of a series of materials with a sulphur concentrate spray. Polyethylene glycol oleate and polyethylene polysulphide were amongst the successful adjuvants, but the majority of the materials he tried were proprietary products of unspecified composition.

The purpose of the present work was to examine representative members of different classes of chemicals which might be incorporated with fungicide sprays as stickers, and to determine the effect of the additives on fungicidal efficiency. In the initial screening test, the effect of the supplement on the tenacity of a cupric oxide deposit on an artificial surface was determined by subjecting the deposit to a standard washing procedure and analysing the residue. The most promising materials from this test were used on leaf surfaces and in field trials, and their influence on the effectiveness of the fungicide was assayed by spore germination.

The physico-chemical factors influencing the tenacity of pesticides alone on different types of surfaces and the influence of supplements on the size of ' run-off ' deposits will be discussed in future communications.

## Experimental

*General procedure for the determination of tenacity*

Suspensions of cupric oxide with different supplements were sprayed, to below ' run-off ', on to cellulosed glass slides (7·6 × 2·5 cm.) or cut leaves. A spray tower was used for preliminary experiments and a spray wheel for the later tests. The spray deposit was dried and then washed with artificial rain, and the proportion of deposit retained by the surface determined chemically or by weighing.

*Materials*

*Copper fungicide.*—Cupric oxide was selected as the standard fungicide because of its availability as a pure compound of fixed composition, and because of its low tenacity compared with those of other copper fungicides tested. It was 97·5% pure and sedimentation analysis, with an Andreasen pipette,[5] showed that 60% (by wt.) of the powder had equivalent Stokes diameters in the range 0–15 $\mu$.

*Slide surface.*—A standard artificial surface was used for most of the laboratory determinations of tenacity. The glass slides were dipped in a 2·5% solution of cellulose acetate in a mixture of acetone (3 vol.) and ethyl lactate (1 vol.), then drained and heated at 80° for 18 h.

*Plant surface.*—The upper surface of the leaves of two-month-old cauliflower (var. Majestic) and 3–4-week-old broad bean (var. Giant Windsor), raised in a cold glasshouse, were sprayed immediately after cutting.

*Supplements.*—The supplements included in the tests are given in Tables I and II; details of their formulation and references to previous workers are given in the footnote to these Tables. Where emulsification of the supplement was necessary, ammonium caseinate, prepared by dissolving 5 g. of casein in 10 g. of ammonia solution (sp. gr. 0·88), centrifuging and making up to 500 ml., was used. This 1% solution was emulsified in a M.S.E. homogenizer with half its volume of the supplement. Preliminary experiments had shown that ammonium caseinate did not reduce the tenacity of cupric oxide as much as other emulsifying agents tested.

*Apparatus and methods*

*Spray tower.*—The preliminary screening tests were carried out with deposits formed in a vertical spray tower. The mechanically-stirred suspension was sprayed from a brass atomizer nozzle, in which the liquid jet was fixed centrally in the air annulus, and centred down a 60-cm. stainless steel cylinder. A cellulosed slide was supported at the base of the tower in a polythene holder which exposed only 4·4 × 2·5 cm. of the slide. The volume of spray that fell on the slide was controlled by varying the period of spraying. Five slides were sprayed with each suspension and dried. Their initial deposit was determined by weighing (for CuO + supplement) and chemical analysis (for CuO) of one of them, whilst the other four were washed for different time periods and analysed in the same manner. The coefficient of variation of the deposits on the five slides from each suspension was less than 8%, although for replicates on different days it could be as high as 20%, possibly because of turbulence effects.[6] It was shown by weighing cupric oxide deposits on 12-mm. cover slips placed at random on a slide that there was no significant difference in deposit over the 4·4 × 2·5 cm. slide area. Suspensions containing 2% cupric oxide were used with this apparatus to give deposits of 100–300 $\mu$g. Cu/cm.$^2$ and the total deposits were weighed to 0·01 mg. The spray tower was discarded in favour of the spray wheel at the end of these tests because spraying the five individual slides took a long time and reproducibility of deposit required constant attention to the atomizer nozzle.

*Spray wheel.*—A truncated cone (angle 65°), 66 cm. diameter, of aluminium was mounted on a horizontal wheel driven at constant speed (12·5 r.p.m.). The slides or leaves, backed on filter paper, were attached to the outside of the cone and their long edges were aligned parallel to the top edge of the cone. The suspension was sprayed from an Aerograph MP paint atomizer directed at right angles to the slides and mounted 30 cm. from the wheel. The slides and leaves were fixed to the filter paper with ' Sellotape ' so as to expose a fixed area, 4·4 × 2·5 cm., to the spray. The cone held a maximum of nine slides leaving the remaining free space to begin and end the spraying. The coefficient of variation between the deposits on the slides in an experiment was always less than 6%; day-to-day variation was as great as with the spray tower. Three of the nine slides were analysed to determine the initial deposit.

## Table I

*Screening tests on the effect of supplements on the tenacity of cupric oxide on cellulose acetate*

| Cupric oxide at 2% Supplement | Concn. g./100 ml. | Ratio of rate of loss of supplement to rate of loss of CuO | Spray tower Tenacity of CuO (× 100) after 10-sec. wash | after 60-sec. wash |
|---|---|---|---|---|
| Nil | | | 24 | 12 |
| *Oils* | | | | |
| Turpentine | 1·0 | 0 | 31 | 14 |
| Castor | 1·0 | 0 | 46 | 32 |
| Cottonseed | 1·0 | 0 | 61 | 42 |
| Linseed | 1·0 | 0 | 81 | 64 |
| Anthracene | 1·0 | 0 | 32 | 21 |
| Liquid paraffin | 1·0 | 0 | 46 | 17 |
| *Gums and resins* | | | | |
| Agar | 0·10 | 0 | 71 | 40 |
| Dextrin | 0·66 | 0 | 15 | 1 |
| Colophony | 1·0 | — | 41 | 22 |
| Canada balsam | 1·0 | — | 33 | 25 |
| *Protein materials* | | | | |
| Gelatin | 0·50 | + | 64 | 25 |
| ,, | 0·05 | + | 16 | 3 |
| Animal protein | 0·50 | 0 | 60 | 43 |
| Casein (soluble) | 0·20 | 0 | 18 | 10 |
| Lime casein (lime : casein, 7 : 1) | 4·0 | + | 70 | 60 |
| *Inorganic salts* | | | | |
| Calcium hydroxide | 1·0 | — | 42 | 5 |
| Aluminium hydroxide | 2·0 | 0 | 49 | 19 |
| *Clays* | | | | |
| *Kaolin | 1·0 | 0 | 17 | 11 |
| Bentonite A | 2·0 | + | 4 | 2 |
| *Synthetic resins* | | | | |
| Coumarone resin | 1·0 | 0 | 99 | 87 |
| Chlorinated polyphenyl | 1·0 | — | 52 | 38 |
| Polyethylene polysulphide | 0·25 | + | 22 | 22 |
| Polyvinyl acetate (PVA) | (0·50 v/v) | 0 | 89 | 55 |
| Polyvinyl chloride (PVC) | (0·50 v/v) | 0 | 89 | 52 |
| *Miscellaneous* | | | | |
| Rubber latex | (0·25 v/v) | + | 94 | 85 |
| Ferrous ammonium alginate | 0·30 | + | 99 | 6 |
| Aluminium stearate | 0·25 | 0 | 19 | 12 |
| Aluminium laurate | 0·25 | + | 36 | 19 |
| Methyl ethyl cellulose | 0·01 | 0 | 32 | 10 |
| Wheat flour | 1·0 | — | 16 | 8 |

* with 1% CuO

+ = ratio > 1; 0 = ratio 1; — = ratio < 1, compared with CuO = 1

*Oils.*[2, 12, 13] The essential oil, turpentine, and representative members of the glyceride and mineral oils were applied as emulsions. Anthracene oil, sp. gr. 1·112, had 4·5% tar acid, and 5·0% tar base content; 67% distilled between 300 and 400°.

*Gums and resins.*[2, 14] Gamboge was triturated with water, whilst colophony and Canada balsam were dissolved in the minimum volume of benzene and emulsified.

*Protein materials.*[1, 2] The animal protein was a dispersible commercial powder used as a sticker in U.S.A.

*Inorganic salts.*[2] The aluminium and ferric hydroxides were precipitated as gels, from potassium aluminium sulphate with aqueous ammonia and from ferric chloride with sodium hydroxide, respectively. The gels were centrifuged and washed.

*Clays.*[3, 15, 16] The kaolin clay was a blended product of low particle size (90% < 2 μ). The bentonites A and B were Wyoming and Redhill grades, respectively. The dispersible bentonite was prepared from bentonite B, aluminium sulphate, and washing soda as recommended by Large.[16]

[*footnote continued under Table II*]

*Washing apparatus.*—After a drying period of 24 h. at 25° for slides, 18 h. for cauliflower leaves, and 3 h. for beans, the deposits were sprayed with a horizontal jet of water. The washing chamber, 60 × 45 cm. and 45 cm. high, was of aluminium sheet with a Perspex top and side, the latter hinged to open outwards. It was slightly inclined to the horizontal to aid drainage. The slide was held vertically by an adjustable metal support between two grooved rods which could be fixed at varying distances from the spray nozzle. Distilled water from an aspirator was fed into a 750-ml. filter flask from which it was forced by compressed air (80 ± 1 cm. Hg) through

**Table II**

*Effect of supplements on the tenacity of cupric oxide on cellulose acetate*

| Cupric oxide at 0·30% | | Spray wheel | |
|---|---|---|---|
| Supplement | Concn. g./100 ml. | Tenacity of CuO (× 100) after 10-sec. wash | after 60-sec. wash |
| Nil | | 8 | 7 |
| *Oils* | | | |
| Tung | 0·30 | 56 | 50 |
| Linseed | 0·30 | 50 | 36 |
| Linseed (boiled) | 0·30 | 57 | 43 |
| Sperm | 0·30 | 23 | 11 |
| *Gums and resins* | | | |
| Agar | 0·30 | 45 | 21 |
| Gamboge | 0·30 | 50 | 30 |
| ,, | 0·15 | 38 | 28 |
| *Inorganic salts* | | | |
| Ferrous sulphate | 0·30 | 18 | 8 |
| Ferric hydroxide | 0·10 | 98 | 65 |
| ,, ,, | 0·03 | 30 | 30 |
| *Clays* | | | |
| Bentonite B | 0·066 | 12 | 8 |
| Dispersible bentonite B | 0·50 | 12 | 11 |
| *Synthetic resins* | | | |
| Polyethylene polysulphide | 0·06 | 20 | 11 |
| Polystyrene | (0·25 v/v) | 35 | 19 |
| Polyvinyl *iso*butyl ether | 0·30 | 32 | 17 |
| *Miscellaneous* | | | |
| Rubber latex | (0·16 v/v) | 69 | 58 |
| Chlorinated rubber | 0·12 | 52 | 34 |
| Ferric ammonium alginate | 0·11 | 52 | 43 |
| Wheat flour + calcium hydroxide | 0·10 + 0·006 | 19 | 10 |
| Methylchlorosilane | 0·01 | 16 | 8 |
| 2-Heptadecylglyoxalidine + calcium hydroxide | 0·50 + 0·05 | 30 | 15 |
| Cetylpyridinium chloride | 0·035 | 23 | 11 |
| Polyethylene glycol mono-oleate | 0·10 | 11 | 10 |

*Synthetic resins.*[3, 4, 14] These were all commercial samples. Coumarone resin (m.p. 36·6°), chlorinated polyphenyl (mixture of *o*-, *m*- and *p*-terphenyl, 60% Cl/wt.), and low viscosity polyvinyl *iso*butyl ether, were dissolved in the minimum amount of benzene and emulsified. The polyethylene polysulphide was a water-dispersible paste with 44% inert carrier. Polystyrene, PVA and PVC were latices prepared by emulsion polymerization. The PVA had been plasticized with dibutyl phthalate and emulsified with polyvinyl alcohol; the latex had 60% solid content and less than 0·3% free monomer.

*Miscellaneous.* The rubber latex[14] was a 60% centrifuged latex containing ammonia as preservative. Chlorinated rubber (10 g.) was dissolved in dibutyl phthalate (10 g.) and carbon tetrachloride (110 g.) and emulsified with an anionic surface-active agent (sodium salt of highly sulphated oil). Ferrous and ferric ammonium alginate solutions were prepared by adding equivalents of ferrous or ferric ammonium sulphate to a solution of low viscosity sodium alginate (equivalent weight 270). A water-dispersible grade of aluminium stearate was used whilst the aluminium laurate was dissolved in benzene and emulsified. The wheat flour was soaked overnight before adding the calcium hydroxide.[17] A 2% solution of methylchlorosilane in carbon tetrachloride was emulsified with a non-ionic surface-active agent (polyethylene glycol type). 2-Heptadecyl-glyoxalidine was a water-miscible liquid containing 54% active material. Cetylpyridinium chloride was above the critical concentration for micelle formation. The polyethylene glycol mono-oleate[4] had an aggregate molecular weight of 600.

a Bray single-hole ceramic nozzle (Type J) to give a horizontal fan-shaped spray with droplets in the range 50–200 $\mu$. The flask was enclosed in a steel box with a Perspex window on the far side, facing a mirror which enabled the level of water in the flask to be adjusted before each wash. A hand-operated shutter controlled the period of spraying.

The apparatus was adjusted to give a uniform spray over an area 2·5 × 4·4 cm. When phials (internal diameter 0·87 cm.) were placed at the slide position facing the spray, the water collected in each phial during a 20-sec. wash was 0·449 g. (S.E. = ± 0·017), and the apparatus gave a 'rainfall' of 0·38 cm./10 sec. with a coefficient of variation of 3·8% (for Table I the 'rainfall' was 0·20 cm./10 sec.). Slides were usually washed for 10, 20, 30 and 60 sec., two fillings of the flask being necessary for the latter wash.

When leaves were washed, an area 2·5 × 4·4 cm., defined by a polythene stencil, was cut from each leaf and these segments were stuck with 'Durofix', on to slides at the spray deposit position.

The tenacity values (× 100) obtained with the wash apparatus replicated to within ± 3 on cellulose acetate, and to within ± 5 on bean and cauliflower. All the tenacity values given are the mean values from at least three determinations, except for Table I where only the results with the best stickers were confirmed by replication.

*Bioassay of deposits.*—A spore germination technique[7] was used to measure the *in vitro* fungistatic activity of spray deposits of cupric oxide, with and without supplements. Glass slides were washed in permanganic acid, rinsed with glass-distilled water, cellulosed, and then sprayed on the spray wheel with the cupric oxide suspensions for a varying number of revolutions to give a dosage range. The test fungus used was a seven-day-old culture of *Alternaria tenuis* (grown on peptone agar), as recommended by previous workers.[8] Spore suspensions of 50,000/ml. were prepared by gently washing the spores from the substrate, centrifuging and washing twice, then suspending in glass-distilled water. Two drops of the suspension were pipetted on to each slide, and the slides placed in a moist chamber and incubated at 25°. After 17 h., the germination of fifty spores in each drop was recorded, and the drop diameter noted if it differed from the mean spreading diameter of 8 mm. Paraffined culture rings were used to contain drops on very readily wettable surfaces;[8] they had no effect on spore germination. The germination on unsprayed slides was 98–100%. The slide deposits were analysed for copper and dosage-mortality data were computed by probit analysis.[9]

*Analytical procedure.*—The initial spray deposits and the washed residues were removed from the slides with hot 1 : 1 nitric acid and the organic matter destroyed by digestion with perchloric acid. The leaves were ashed and the residue dissolved in nitric acid. Copper was determined colorimetrically with sodium diethyldithiocarbamate in 0·1% gelatin.[10] With bioassay deposits of less than 10 $\mu$g. of copper, the copper was extracted with carbon tetrachloride as the diethyldithiocarbamate complex, in the presence of ethylenediaminetetra-acetic acid.[11]

## Results

*Screening test for supplements*

Table I gives the tenacity results with deposits formed in the spray tower, after washing for 10 and 60 sec. Since the slides were weighed and the deposits also analysed for copper, the relative rate of loss of supplement to cupric oxide could be calculated. Values of this ratio greater than, equal to or less than unity are shown in Table I by the symbols +, 0, and −, respectively.

The data show that most of the supplements tested gave only a small increase in the tenacity of cupric oxide and that only linseed oil, agar, lime casein, the synthetic resins (coumarone, polyvinyl acetate [PVA] and chloride [PVC]), and rubber latex merited further investigation. A number of commercial stickers were included in the test but none of them gave more tenacious deposits than those given above and the results are not given (with the exception of dispersible animal protein).

Fig. 1 shows some of the most widely different tenacity curves obtained. For most of the supplements with cupric oxide, however, the general form of the curve was similar to that of cupric oxide alone.

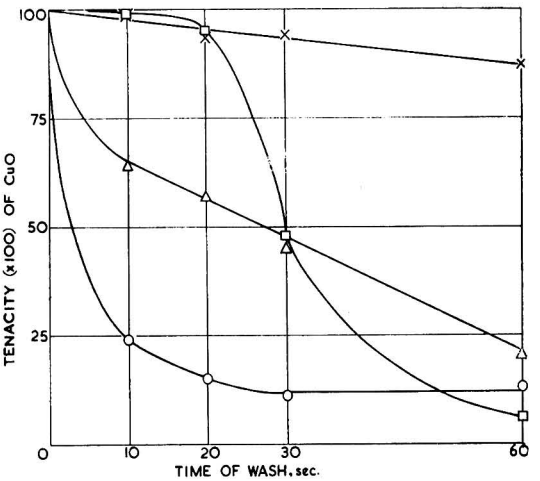


FIG. 1.—*Tenacity of cupric oxide alone, and with supplements, on cellulose acetate*

○ CuO
□ CuO + ferrous ammonium alginate
× CuO + coumarone resin
△ CuO + gelatin (0·5%)

Table II gives the corresponding tenacity values obtained after spraying on the wheel. Strict comparisons between Tables I and II are not possible since the initial deposits are much lower for Table II (8–15 $\mu$g. Cu/cm.$^2$) and the washing intensity was more severe. However, where the same supplements (linseed, agar, rubber latex and polyethylene polysulphide) were included in both series of tests, the same relative behaviour was shown.

Table II confirms the preliminary conclusion that the vegetable glyceride oils, especially the drying type, are effective stickers and shows that the faster-drying boiled linseed oil is a more efficient sticker than the raw oil. The concentrations of gamboge and ferric hydroxide necessary for high tenacities give solutions too viscous to be practicable. The chlorinated rubber emulsion was also difficult to spray because it instantly flocculated the cupric oxide into large agglomerates. The ferric ammonium alginate, which became insoluble on drying, was promising but, because of the viscosity of the solution, gave low initial deposits.

For comparison with Table II, the tenacity (× 100) of the copper present in a standard 10 : 12·5 : 100 Bordeaux mixture [lb. $CuSO_4,5H_2O$ : lb. $Ca(OH)_2$ : gal. water], for 11 $\mu$g. Cu/cm.$^2$ initial deposit, was 94 and 84, for 10- and 60-sec. washes, respectively.

*Tenacity of 0·3% cupric oxide with boiled linseed oil, PVA and rubber latex*

The effect of varying the CuO : supplement ratio on the tenacity of a deposit on cellulose acetate was determined with the supplements, boiled linseed oil, PVA and rubber latex, each sprayed at various concentrations with 0·3% cupric oxide. These tests were repeated on freshly-cut bean leaves to determine the influence of the surface on this behaviour. Preliminary experiments had shown that the tenacity of cupric oxide on freshly-cut leaves did not differ from the tenacity of a similar leaf deposit which had dried on the plant before it was washed.

All the deposits were analysed chemically. Initial deposits varied from 8 to 10 $\mu$g. Cu/cm.$^2$. The tenacities on cellulose acetate after a 60-sec. wash are given in Fig. 2 and those on leaves in Fig. 3. The 1% v/v rubber latex agglomerated the cupric oxide and made spraying difficult; this concentration was not used on beans. The supplement concentration had an important effect on the tenacity of the deposit. Figs. 2 and 3 indicate that the optimum concentrations are independent of the cellulose acetate and bean-leaf surfaces used.

Table III gives the tenacity of cupric oxide with the supplements, at their minimum effective concentrations (as shown in Figs. 2 and 3), on cauliflower leaves.

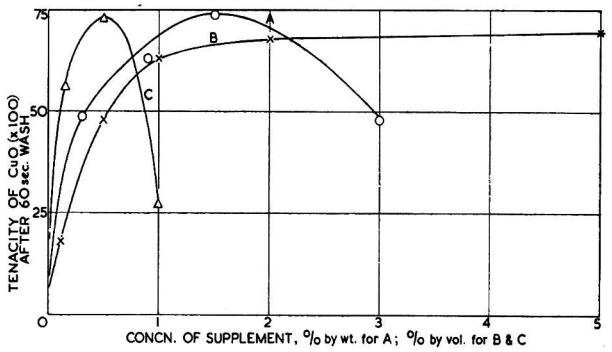


FIG. 2.—*Effect of varying supplement concentration on the tenacity of cupric oxide* (0·3%) *on cellulose acetate*

A ○ Boiled linseed oil
B × PVA
C △ Rubber latex

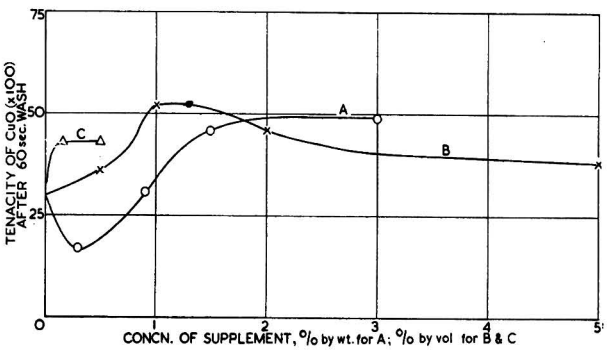


FIG. 3.—*Effect of varying supplement concentration on the tenacity of cupric oxide* (0·3%) *on bean leaves*

A ○ Boiled linseed oil
B × PVA
C △ Rubber latex

## Table III

*Tenacity of cupric oxide with and without supplements on cauliflower leaves* (0·3% *cupric oxide*)

| Supplement | Tenacity of CuO (× 100) after 60-sec. wash |
|---|---|
| Nil | 15 |
| Boiled linseed oil (1·5%) | 63 |
| PVA (2% v/v) | 82 |
| Rubber latex (0·5% v/v) | 79 |

*Effect of supplements on the particle size distribution of cupric oxide suspensions*

The sedimentation analysis of 0·3% cupric oxide suspensions with boiled linseed oil, PVA and rubber latex was determined with an Andreasen pipette[5] at 25·0 ± 0·1°. The cupric oxide contents of the pipetted samples were analysed chemically. The curves for the weight per cent cupric oxide under size at each level of Stokes' diameter ($d_s$) are given in Fig. 4. The corresponding curves for cupric oxide alone and with a wetting agent (sodium dinonyl sulphosuccinate) are included for reference. The $d_s$ values for PVA latices were calculated using viscosity data obtained with a modified Ostwald viscometer. Because of the density of the cupric oxide, it was only possible to observe the behaviour of particles with equivalent $d_s$ values below 25 $\mu$.

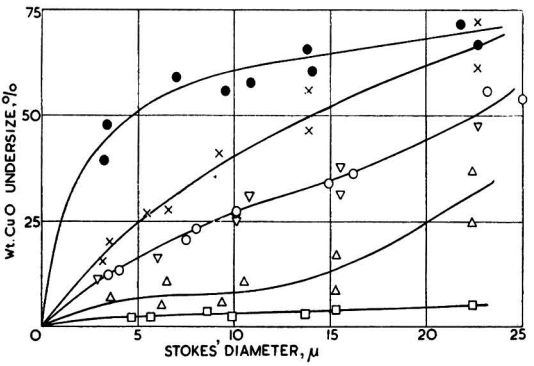


FIG. 4.—*Particle size distribution curves for cupric oxide* (0·3%), *alone and with supplements*

○ CuO alone
△ CuO + 0·5% v/v rubber latex
× CuO + 2·0% v/v PVA
● CuO + 0·1% sodium dinonyl sulphosuccinate
□ CuO + 0·3% boiled linseed oil
▽ CuO + 1·0% v/v PVA

The results show that, whereas 1% v/v PVA did not affect the dispersibility of the cupric oxide, 2% v/v PVA gave a considerable improvement. The rubber latex and boiled linseed oil flocculated the suspensions. Microscopical examination of the linseed oil–CuO system showed that the emulsion broke quickly and the cupric oxide was flocculated as oil-wetted agglomerates. When boiled linseed oil was replaced by the raw oil a similar effect was obtained.

## Field trials

In 1953, field trials were carried out on potato and tomato foliage sprayed with cupric oxide and some of the most successful stickers from the preliminary screening test (Table I). In both trials, Bordeaux mixture was included as a reference fungicide. The tenacity of copper residues on the foliage was determined chemically, and with potato the control afforded by the treatments against leaf destruction by blight (*Phytophthora infestans*) was used as a measure of their fungicidal efficiency.

### *Experimental*

#### (*a*) *Potato foliage*

A plot of 500 King Edward potato plants was divided into five randomized blocks each consisting of ten individual plots of ten plants, and to each of these plots one of the ten treatments was applied.

*Spray treatments.*—These were made with the following mixtures:

Control
0·086% CuO (≡ 0·066% Cu)
do. + 0·033% agar
do. + 0·086% linseed oil
do. + 0·60% calcium hydroxide + 0·086% casein
do. + 0·083% v/v PVA (i.e. 0·67 pt./100 gal.)
do. + 0·086% coumarone resin
do. + 0·041% v/v rubber latex (i.e. 0·33 pt./100 gal.)
do. + 0·083% v/v PVC (i.e. 0·67 pt./100 gal.)
Bordeaux mixture, $CuSO_4,5H_2O$ 4 lb., $Ca(OH)_2$ 6 lb., water 150 gal. (≡0·066% Cu).

*Spray application.*—One and a half gal. of the spray suspension was applied to each individual plot with a watering-can (except for the agar and Bordeaux treatments which required a hand-pump spray). This ensured complete wetting of all foliage beyond 'run-off' and negligible

spray drift. Since growth was rapid, two spray applications were necessary to combat blight; they were made on 19 July and 10 August.

*Sampling and analysis of the spray residue.*—The leaves were sampled for the initial deposits as soon as the deposits had dried. The plants were sampled at mid-height to avoid the new growth at the top and the old leaves at the bottom; 20 leaflets were collected at random from each plot. A 1·73-cm. disc was cut with a stainless steel cork borer from the centre of a bundle of these leaflets, avoiding the leaf tips and edges. The samples were ashed and analysed for copper. A copper blank on unsprayed leaves was determined at each sampling.

*Blight assessment.*—The incidence of potato blight on each individual plot was estimated with the British Mycological Society assessment key.[18]

(*b*) *Tomato foliage*

A plot of 200 tomato plants (var. Market King) was divided into four randomized blocks, each consisting of five plots of ten tomatoes, and to each of these plots one of the five spray treatments was applied.

*Spray treatments.*—These were made using the following mixtures:

Control
0·086% CuO (≡ 0·066% Cu)
do. + 0·086% linseed oil
do. + 0·041% v/v rubber latex (i.e. 0·33 pt./100 gal.)
Bordeaux mixture, $CuSO_4,5H_2O$ 4 lb., $Ca(OH)_2$ 6 lb., water 150 gal. (≡ 0·066% Cu).

Only one spraying was made (28 July). The spray application and sampling procedure were as for potatoes, but as there were fewer leaves from which to sample, ten were taken from each plot, i.e. one per plant, choosing the top leaves.

*Results*

The initial deposits and residues of copper from the spray treatments applied to potato and tomato foliage are given in Tables IV and V, respectively. The limits given are approximate 95% confidence limits for each mean quoted, i.e. ± 2 S.E.

**Table IV**

*CuO and Bordeaux residues on potato foliage, 1953 (in mg. Cu/100 sq. cm. of leaf)*

| Date of sampling | Spray treatment | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | CuO | CuO + Supplement | | | | | | | Bordeaux |
| | | Agar | Linseed oil | Lime casein | PVA | Coumarone resin | Rubber latex | PVC | |
| 19 July | 1·00 ± 0·34 | 1·19 ± 0·08 | 1·55 ± 0·38 | 0·76 ± 0·19 | 0·70 ± 0·15 | 0·81 ± 0·11 | 0·89 ± 0·17 | 1·00 ± 0·30 | 1·15 ± 0·23 |
| 6 August | 0·02 ± 0·04 | 0·25 ± 0·13 | 0·28 ± 0·28 | 0·28 ± 0·23 | 0·23 ± 0·13 | 0·06 ± 0·04 | 0·08 ± 0·06 | 0·13 ± 0·06 | 0·56 ± 0·15 |
| 10 August | 1·08 ± 0·17 | 1·56 ± 0·66 | 1·20 ± 0·23 | 0·70 ± 0·04 | 1·68 ± 0·70 | 1·62 ± 0·45 | 1·05 ± 0·49 | 1·05 ± 0·21 | 1·24 ± 0·34 |
| 19 August | 0·17 ± 0·06 | 0·55 ± 0·23 | 0·51 ± 0·08 | 0·32 ± 0·11 | 0·28 ± 0·11 | 0·51 ± 0·25 | 0·15 ± 0·06 | 0·36 ± 0·15 | 0·89 ± 0·17 |
| 27 August | 0·02 ± 0·02 | 0·36 ± 0·15 | 0·25 ± 0·08 | 0·13 ± 0·08 | 0·13 ± 0·04 | 0·15 ± 0·06 | 0·04 ± 0·04 | 0·25 ± 0·08 | 0·60 ± 0·06 |

*Effect of supplements on initial deposits.*—The initial deposits of copper on potato and tomato leaves were analysed statistically. In both trials the only significant increase in initial deposit given by the supplements, when compared with cupric oxide alone, was that given by linseed oil—

significant at the 1% level for the first application on potato, and at the 0·1% level on tomato. The data for the second application on potato are complicated by the residues from the first application, still resident on the leaf surface.

*Effect of supplements on tenacity.*—The tenacities in Table VI give the amount of copper residue at the sampling date as a percentage of the initial deposit at the last spray application. These percentages were transformed to ' angles of equal information ' and analysed statistically ; the significant differences of the treatment tenacities from the tenacity of cupric oxide alone are starred in Table VI. The rapid growth of potato between the first spray application and sampling was probably responsible for the low tenacities on 6 August ; as there was little new growth after the second application, the later data are more reliable. On potato all the spray supplements, with the exception of rubber latex, improved the tenacity of cupric oxide. Coumarone resin and PVA gave only a small increase in tenacity but agar, linseed oil, lime casein and PVC gave appreciably improved tenacities. There were no differences (at the 5% level) between the last four treatments.

## Table V

*CuO and Bordeaux residues on tomato foliage, 1953 (in mg. Cu/100 sq. cm. of leaf)*

| Date of sampling | Spray treatment | | | |
|---|---|---|---|---|
| | CuO | CuO + linseed oil | CuO + rubber latex | Bordeaux |
| 28 July | 1·30 ± 0·45 | 3·99 ± 0·74 | 2·08 ± 0·79 | 1·40 ± 0·21 |
| 14 August | 0·72 ± 0·38 | 1·06 ± 0·36 | 0·64 ± 0·23 | 0·70 ± 0·11 |
| 25 August | 0·06 ± 0·06 | 0·38 ± 0·13 | 0·04 ± 0·04 | 0·21 ± 0·08 |
| 2 Sept. | 0·00 | 0·25 ± 0·17 | 0·02 ± 0·02 | 0·11 ± 0·05 |

## Table VI

*Tenacity (× 100) of the copper residues*
*(% of Cu deposit at last spraying present at sampling date)*

*(a) Potato*

| Supplement | Date of sampling | | |
|---|---|---|---|
| | 6 August | 19 August | 27 August |
| Nil | 3 | 16 | 3 |
| Agar | 21* | 36* | 23*** |
| Linseed oil | 18 | 42** | 21*** |
| Lime casein | 36*** | 44*** | 19*** |
| PVA | 33** | 17 | 8* |
| Coumarone resin | 7 | 32* | 9* |
| Rubber latex | 9 | 14 | 3 |
| PVC | 12 | 34* | 23*** |
| (Bordeaux) | 49*** | 72*** | 51*** |
| Rainfall (in.) | 19 July–6 Aug. = 1·42 | 6–19 Aug. = 0·69 | 19–27 Aug. = 2·71 |

*(b) Tomato*

| Supplement | Date of sampling | | |
|---|---|---|---|
| | 14 August | 25 August | 2 Sept. |
| Nil | 56 | 5 | 0 |
| Linseed oil | 26 | 10 | 6** |
| Rubber latex | 31 | 2 | 1 |
| (Bordeaux) | 50 | 15* | 8** |
| Rainfall (in.) | 28 July–14 Aug. = 0·98 | 14–25 Aug. = 3·40 | 25 Aug.–2 Sept. = 2·44 |

*** Significant at P = 0·001
** ,, ,, P = 0·01
* ,, ,, P = 0·05

Strict comparison between the cupric oxide treatments and Bordeaux are not justified, for the cupric oxide was selected for these tests because of its low tenacity. Whilst Bordeaux gave higher tenacities on potato than any cupric oxide treatment, its tenacity on tomato did not differ (at the 5% level) from that of the linseed oil treatment.

*Effect of supplements on control of blight on potato.*—Fig. 5 shows the progress of blight on potato foliage treated with cupric oxide, alone and with agar, and also with Bordeaux. Statistical analysis of the results, after they had been transformed as above, showed that, whilst the extent of control obtained with cupric oxide was improved (at the 5% level) by the addition of agar, for the mid-points of the curves, none of the other supplements gave an equivalent improvement. All the cupric oxide treatments gave greater blight control (at the 0·1% level) than the unsprayed block, but much less (below the 5% level) than Bordeaux.

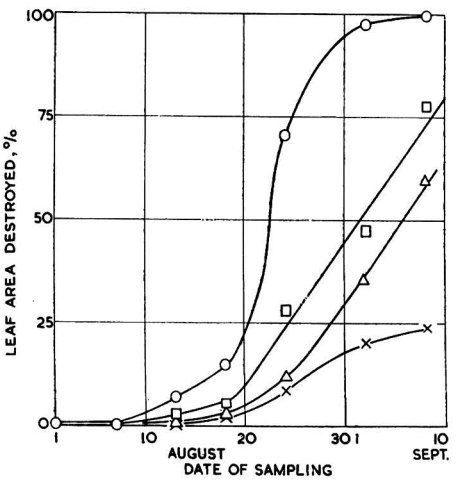


FIG. 5.—*Potato blight progress curves, 1953*

○ Unsprayed
□ CuO
△ CuO + agar
× Bordeaux

*Bioassay of cupric oxide-supplement spray deposits*

The effect of the supplements used in the potato field trials on the fungitoxicity of cupric oxide to *Alternaria tenuis* has been determined. The CuO : supplement ratio was the same as for the field trial (except for PVA and rubber latex), but the absolute concentrations had to be varied to give dosage–response plots. Probit analysis could not be applied to the results with the lime casein treatment, for, when combined with cupric oxide, which gave 1–95% inhibition of spore germination with $3{\cdot}5$–$22{\cdot}5 \times 10^{-5}$ $\mu$g. of Cu/spore, counts of 100% germination were recorded over the same dosage range, i.e. the fungitoxicity of the cupric oxide had been completely inhibited. The probit regression lines for the linseed oil, coumarone resin, and PVC treatments were parallel and the relative potencies of the treatments were calculated. The wide range of the fiducial limits is due to heterogeneity in the response data. Table VII (*a*) shows that whereas the agar, linseed oil and coumarone resin inhibited, PVC increased, the fungistatic activity of the cupric oxide. Neither the benzene solvent for coumarone resin nor the trace of ammonium caseinate solution, used to emulsify the resin and linseed oil, affected these results.

When PVA and rubber latex were examined at a later date, the fungus produced a less resistant and better sporulating strain. Earlier experiments have shown the increase in tenacity conferred on 0·3% cupric oxide by 1 and 2% v/v PVA and 0·5% v/v rubber latex, all giving lower CuO : supplement ratios than were used in the field trial. Table VII (*b*) shows the effect of these lower ratios of supplements on the fungitoxicity of cupric oxide ; the concentrations used were less than those of the tenacity experiments so as to give dosage–response plots, and their equivalent concentrations with 0·3% cupric oxide are recorded. These higher concentrations of supplements produced only slight or no inhibition in the activity of the fungicide.

## Discussion

Further work will be necessary to determine the applicability of the results of these experiments to other fungicides, but the screening test with the supplements on cellulose acetate showed that few of the 47 materials examined gave a substantial increase in the tenacity of a cupric oxide deposit. The artificial rain was sufficiently severe to avoid rating too highly the tenacities of such supplements as ferrous ammonium alginate, which peel off on prolonged washing. The data provide no evidence to support Horsfall's suggestion[19] that the tenacity of a deposit is proportional to the logarithm of the rainfall ; even within the same chemical group supplements behaved in different ways.

**Table VII**

*Dosage–response data: cupric oxide with and without supplements tested against* Alternaria tenuis

| Compound | Equation of line | $ED_{50}$ ($\times 10^{-5}$ µg. Cu/spore) | Relative potency | 95% fiducial limits |
|---|---|---|---|---|
| (*a*) | | | | |
| CuO | $y = 1{\cdot}07x + 3{\cdot}38$ * (0·91–1·23) | 32·7 | + | + |
| CuO + agar | $y = 1{\cdot}57x + 1{\cdot}58$ * (1·27–1·86) | 152 | + | + |
| CuO | $y = 3{\cdot}06x + 2{\cdot}18$ | 83·6 | 1·00 | — |
| CuO + linseed oil | $y = 3{\cdot}06x + 1{\cdot}21$ | 173 | 0·47 | 0·31–0·70 |
| CuO | $y = 2{\cdot}60x + 2{\cdot}82$ | 69·0 | 1·00 | — |
| CuO + coumarone resin | $y = 2{\cdot}60x + 1{\cdot}39$ | 245 | 0·23 | 0·12–0·45 |
| CuO | $y = 2{\cdot}08x + 3{\cdot}31$ | 65·0 | 1·00 | — |
| CuO + PVC | $y = 2{\cdot}08x + 3{\cdot}80$ | 37·8 | 1·75 | 0·95–3·24 |
| (*b*) | | | | |
| CuO | $y = 4{\cdot}48x + 2{\cdot}80$ | 3·09 | 1·00 | — |
| CuO + PVA (≡ 1% v/v) | $y = 4{\cdot}48x + 3{\cdot}32$ | 2·37 | 1·28 | 0·84–1·95 |
| CuO | $y = 5{\cdot}77x + 2{\cdot}06$ | 3·23 | 1·00 | — |
| CuO + PVA (≡ 2% v/v) | $y = 5{\cdot}77x + 1{\cdot}86$ | 3·50 | 0·92 | 0·74–1·15 |
| CuO | $y = 5{\cdot}22x + 2{\cdot}35$ | 3·24 | 1·00 | — |
| CuO + rubber latex (≡ 0·5% v/v) | $y = 5{\cdot}22x + 1{\cdot}57$ | 4·54 | 0·73 | 0·49–1·10 |

* 95% limits of the slope
\+ Dosage–response lines not parallel

Nearly all the best stickers in Table I washed off the slide surface at the same rate as the cupric oxide was removed, which suggests that the supplements had formed a homogeneous complex with the oxide rather than coating the deposit. The most promising supplements in Tables I and II, the synthetic resins and the drying-type glyceride oils, formed coherent spray deposits sufficiently plastic and tenacious not to flake from the slide. Figs. 2 and 3 indicate that although the most effective concentrations of these stickers may be independent of the nature of the surface, the surface has a considerable effect on the increase in tenacity due to the supplement. This suggests that it may be necessary to test on each particular crop the promising materials which pass this screening test on cellulose acetate and do not inhibit the fungicide in the laboratory bioassay, and so determine the specific sticker and its minimum effective concentration.

An important result of the field trials was the preferential retention of cupric oxide when incorporated with linseed oil. On tomato plants, a residue level was obtained which was greater, at all times, than that from a Bordeaux mixture of equivalent copper content. It is the flocculation of the cupric oxide by the linseed oil emulsion that gives the preferentially retained oil-wetted agglomerates. Chemical reaction between emulsifier and solid has been suggested[12, 20] as the cause of this flocculation, but the increase of free surface area alone on the addition of the solid could so denude the emulsion of surface-active agent that the emulsion breaks. Unfortunately, special spray equipment is required[21] for the practical application of these emulsions.

The increase in cupric oxide tenacity on potato plants given by the supplements did not lead to an equivalent increase in blight control. Laboratory bioassay has shown that agar, coumarone resin and linseed oil lower the fungitoxicity of cupric oxide whilst lime casein produces complete inhibition. Green & Goldsworthy[22] ascribed the poor fungus control with a glyceride oil supplement to the formation of an impervious coating on the fungicide which prevents the diffusion of copper to the fungus spores, but the flocculation of cupric oxide by the linseed oil emulsion (Fig. 4) could almost equally be responsible, since the decrease in fungitoxicity with increasing particle size is well established.[19] Conversely, the increasing fungitoxicity with PVC could be

due to its surface-active agent content increasing the dispersibility of the cupric oxide, a method of increasing fungicidal activity which does not appear to have been previously investigated. With lime casein the protein competes with the spores for copper.[23] The concentrations of PVA and rubber latex in the laboratory bioassay were equivalently greater than the concentrations used in the field trial but even these interfered little with the biological effectiveness of the fungicide. The slight decrease with rubber latex, shown in Table VII (*b*), could be due to its flocculation of cupric oxide suspensions, as above.

The general conclusion from this work is that, of the selected materials from the screening test, only the latex-type of synthetic resins, such as PVA, gave promise as stickers that did not reduce the potency of the cupric oxide fungicide. The poor performance of PVA in the field trial was probably due to the low concentration at which it was used, since laboratory tests on different surfaces showed its efficiency at higher concentrations. It is proposed to carry out further experiments with PVA as a sticker for copper fungicides at these higher concentrations.

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JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE

# ABSTRACTS

FEBRUARY, 1956

The general arrangement of the abstracts is as follows: 1.—AGRICULTURE AND HORTICULTURE. 2.—FOOD; also appropriate Microbiological Processes; Essential Oils. 3.—SANITATION, including Water; Sewage; Atmospheric Pollution, etc. 4.—APPARATUS AND UNCLASSIFIED.

## INDEX OF AUTHORS' NAMES

JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE

# ABSTRACTS

FEBRUARY, 1956

## I.—AGRICULTURE AND HORTICULTURE

### General : Soils and Fertilisers

**Meaning of the great German soil fertility survey.—A reply to criticism.** F. van der Paauw (*Soil Sci.*, 1955, **80**, 253—254).—The criticism by Willcox (*Soil Sci.*, 1955, **79**, 123—132) is answered. T. G. MORRIS.

**Soils of Zanzibar Protectorate.** W. E. Calton, G. E. Tidbury and G. F. Walker (*E. Afr. agric. J.*, 1955, **21**, 53—60).—Physical, chemical and mineralogical characteristics of the important soils of the two main islands of the Zanzibar Protectorate are presented. Soil utilisation in relation to soil type is also discussed. A. H. CORNFIELD.

**Arctic brown soil.** J. C. F. Tedrow and D. E. Hill (*Soil Sci.*, 1955, **80**, 265—275).—The name Arctic brown soil is suggested for the active layer (that which thaws in the summer) overlying the permanently frozen soil. Well drained Arctic brown soil has characteristics different from those of wet tundra. It forms under a cover of heaths, herbs, sedges, grasses and lichens. The relation of this soil to the landscape, its morphology, mechanical composition and genesis are discussed. T. G. MORRIS.

**Basalt soils in Britain and Australia.** A. B. Costin (*J. Soil Sci.*, 1955, **6**, 268—269).—Characteristics of basaltic soils in Britain are compared briefly with those formed from similar material in Australia. A. H. CORNFIELD.

**Soils of Nuits-Saint-Georges.** B. Bel (*Ann. Agron.*, 1955, **6**, 385—421).—The geography, geology, pedology and agronomic characteristics of the region are described. A. H. CORNFIELD.

**Characteristics of the soils of the Beauce region.** J. Dupuis and A. Cailleux (*Ann. Agron.*, 1955, **6**, 373—383).—Geological and pedological characteristics of the region are described. A. H. CORNFIELD.

**Soil minerals of the Gold Coast.** R. Hamilton (*J. Soil Sci.*, 1955, **6**, 312—318).—The mineralogical constitution of five groups of dark-coloured soils representing the melanites (alkaline, black, heavy clays) and one alluvial fluvio-marine soil (alluvite) are presented. A. H. CORNFIELD.

**Source and characteristics of mineral materials from which South Carolina coastal plain marine terrace soils are derived.** H. P. Cooper (*Soil Sci.*, 1955, **80**, 221—228).—The sources of the soil materials are discussed, and the special characteristics of the different river basins are described. T. G. MORRIS.

**Profile development in the sand dunes of Culbin Forest, Morayshire. I. Physical properties.** T. W. Wright (*J. Soil Sci.*, 1955, **6**, 270—283).—Changes in some physical properties of sand dunes brought about by fixation (with brushwood thatching) and afforestation on sites ranging from bare sand to mature plantations are reported. A. H. CORNFIELD.

**Grey wooded (podsolic) soils of Saskatchewan, Canada.** H. C. Moss and R. J. St. Arnaud (*J. Soil Sci.*, 1955, **6**, 293—310).—A review of past work is given and results of recent work on the chemical and morphological characteristics of grey wooded soils of western Canada are presented. A no. of varieties or types of these soils have been recognised. The problems of classification and nomenclature of the soils are discussed. A. H. CORNFIELD.

**Physical and chemical conditions in Danish marsh land.** H. C. Aslyng (*Tidsskr. Planteavl.*, 1955, **59**, 328—344).—Marine-deposited clays on the west coast of the Jutland peninsula are described. Heavy applications of superphosphate or gypsum improved the structure and increased infiltration rates somewhat, the reduction in Na content corresponding with about ⅓ of the Ca applied. A. G. POLLARD.

**Crop production on former vineyard soils.** L. Depardon and P. Buron (*Ann. Agron.*, 1955, **6**, 161—167).—The physical and chemical properties of former vineyard soils and comparable agricultural soils and the composition of crops grown on the two types are presented. The vineyard soils were usually poorer in N but much richer in Cu (due to application of Cu fungicides to the vines) and had a lower pH than had the agricultural soils. Vineyard soils were not lacking in P, K, Mg or Mn. A. H. CORNFIELD.

**Titania content of some grey-brown podsolic soils of East Pakistan.** A. Karim and D. H. Khan (*Soil Sci.*, 1955, **80**, 277—280).—The profile distribution of $TiO_2$ in the whole soil and in the colloid fractions of some grey-brown soils is reported. In all but one of the soils examined, the $TiO_2$ distribution follows the same trend as that of the clay fraction. In the profile of the whole soil $TiO_2$ moves downward and accumulates in the B horizon. T. G. MORRIS.

**Leaf leachates as a factor in pedogenesis.** C. Bloomfield (*J. Sci. Food Agric.*, 1955, **6**, 641—651).—The mechanism of formation of two soil types widely occurring in the northern hemisphere, viz., gley and podsolic soils, is discussed. Fe reduction and migration are generally considered to be caused by the action of the humose upper horizons normally present in these soils. In tests water extracts of unhumified plant debris, i.e., of various coniferous and broadleaved trees caused relatively extensive solution of sesquioxides, $Fe^{...}$ being reduced in the process. Raising the pH and aëration of the extracts decreased the extent of solution and reduction. A mechanism is postulated for the formation of the $A_2$ horizon of a podsolised soil in terms of the ability of the material leached from fallen leaves by percolating rain water to dissolve, reduce and form a complex with sesquioxides. (27 references.) E. M. J.

**Genesis of clay minerals in soil. II. Soil derived from granodiorite at Ogoe, Fukushima Prefecture, Japan.** Jun-ichi Masui (*Tohoku J. agric. Res.*, 1954, **5**, 71—91).—Minerals in these soils are examined by *X*-ray powder patterns and by chemical and thermal analyses. Nontronite, montmorillonite and halloysite were the principal minerals; some degraded illite and kaolinite were also present. The mechanism of the formation of these minerals is discussed. A. G. POLLARD.

**Some clay minerals in paddy soils.** N. Uchiyama and Y. Onikura (*Tohoku J. agric. Res.*, 1954, **5**, 159—176).—Minerals separated from an acid paddy soil are examined chemically, by differential thermal analysis and by *X*-diffraction methods. Data presented demonstrate the presence of considerable proportions of an expanding, 2 : 1-lattice clay and stratified mixed-layer minerals having varied and large basal spacings. These are possibly secondary products of which the mechanism of formation is not clear. A. G. POLLARD.

**Reaction between bentonite and certain naturally-occurring compounds.** E. R. Turner (*J. Soil Sci.*, 1955, **6**, 319—326).—Excretions from clover roots formed a blue colour with bentonite. No colour was formed with mica or illite. The depth of colour and the % of plants giving the colour with bentonite varied among different lines of clover. Colour formation also occurred with other, non-leguminous, species. Adsorption of compounds in the crystal lattice may be an important step in the formation of colour. The colour reaction between bentonite and root excretions from clover resembled that between bentonite and indolyl-derivatives. Fe present in the lattice or as an impurity in bentonite is an important factor in colour development. A. H. CORNFIELD.

**Podsolisation. VI. Immobilisation of iron and aluminium.** C. Bloomfield (*J. Soil Sci.*, 1955, **6**, 284—292).—The sorption of Fe- and Al-leaf leachate compounds on soil colloids was studied. In general, the extent of sorption varied inversely as the efficiency of the species as a podsolising agent. A. H. CORNFIELD.

**Coarse particle distribution in the skeleton of some coarse- to medium-textured soils.** C. L. W. Swanson, A. Ritchie, jun., and H. A. Doehne (*J. Soil Sci.*, 1955, **6**, 209—218).—The distribution of coarse mineral particles (>3 in., 3—2 in., etc., down to 5—2 mm.) through the profile of a no. of soils is presented. The effects of coarse particle size on losses by erosion, gross physical properties, and on the classification of soils are discussed. A. H. CORNFIELD.

**Effect of electrolyte concentration on soil permeability.** J. P. Quirk and R. K. Schofield (*J. Soil Sci.*, 1955, **6**, 163—178).—The permeability of a silty loam soil to successively decreasing concn. of the Cl′ of Na, K, Mg and Ca as well as to mixed chlorides containing varying ratios of Na : K and with varying degrees of Na saturation of the soil was studied under laboratory conditions. Decrease in the permeability of the soil below a certain concn. of percolating solution, which was sp. for each cation, occurred. This "threshhold concn." decreased in the order Na, K, Mg and Ca. The threshold concn. of Ca in the mixed Ca–Na system was the same, at all levels of soil Na saturation (6—35%), as when only Ca was

present in the system, in spite of the fact that the threshold concn. of Na increased with the % of Na saturation of the soil. It should be possible to maintain the permeability of an irrigated soil irrespective of its degree of Na saturation providing a sufficient concn. of electrolyte is present. The concn. can be increased, if necessary, by addition of sol. Ca salts to the irrigation water. A. H. CORNFIELD.

**Improvements in the structural state of soils under leys.** A. J. Low (*J. Soil Sci.*, 1955, **6**, 179—199).—The rate of physical improvement, as measured by field observations and water-stable aggregation, of old arable soils sown to leys was studied. The rate of improvement was greater when a high than when a low state of water-stable aggregation existed at the time of sowing the leys. Poor rate of improvement occurred if fields were closely grazed and compacted, whilst high aggregation often occurred just after a hay crop or after a period of vigorous growth. Rate of structural improvement was better in well- than in poorly-drained soil. Structural changes took place more rapidly in soils of either high coarse sand or clay content than in those of medium clay and high fine sand and/or silt content. On clay loams 50 years or more of leys may be necessary to restore the physical condition of arable soil to that of old grassland, whilst on sandy loams 5—10 years may be sufficient. A. H. CORNFIELD.

**Water sorption and swelling of clay blocks.** J. W. Holmes (*J. Soil Sci.*, 1955, **6**, 200—208).—The p*F*-moisture content curves during wetting and drying of blocks of moulded clay obtained from red-brown earth and black earth soils (predominantly illitic and kaolinitic) over the tension range p*F* 2·0—6·0, was studied. Changes in pore space due to swelling were more important than filling or emptying of the pore space by water in determining total water uptake. The p*F*-moisture content curves of natural clay aggregates were very similar to those of moulded blocks. The possible causes of hysteresis in the wetting-drying cycle are discussed. A. H. CORNFIELD.

**p*F*-water relationships and pore size distribution in Delhi soil and Jumna sand.** K. Subba Rao, P. T. Ramacharlu and P. S. Talwar (*J. Indian Soc. Soil Sci.*, 1955, **3**, 1—6).—p*F*-water content curves from p*F* 0—3 during the drying of Delhi (Gangetic alluvial) soil and Jumna river sand are presented. The two curves were distinctly different. At 500 cm. tension only 1% of the total pore vol. in the sand and 31% of that in the soil was filled with water. In the soil nearly 60% of the total pore vol. consisted of pores <0·02 mm. diameter, whilst in the sand approx. 60% of the pore space consisted of pores 0·2—0·05 mm. diameter. The permeability coeff. (for water) of the soil was 5·6 and of the sand 143·0 cm. per hr. A. H. CORNFIELD.

**Soil fertility: a watershed management problem in the San Gabriel mountains of Southern California.** H. Hellmers, J. F. Bonner and J. M. Kelleher (*Soil Sci.*, 1955, **80**, 189—197).—Laboratory tests have shown that the native vegetation of this area responded significantly to fertilisation with N alone or in combination with P, but trace elements and other nutrients were in sufficient amount. Attempts to increase natural vegetation by manuring as a means of minimising wind and water erosion are recorded. T. G. MORRIS.

**Balance of water in soils: relationship between precipitation, evaporation and run-off.** L. Turc (*Ann. Agron.*, 1955, **6**, 5—131).—A no. of formulæ for calculating the balance of water in soils are presented and discussed. Uses of the formulæ are illustrated by applying them to results of a variety of lysimeter experiments. A. H. CORNFIELD.

**Thermodynamics of soil moisture: a new application.** K. L. Babcock and R. Overstreet (*Soil Sci.*, 1955, **80**, 257—263).—The basic assumptions involved in applying thermodynamics to the consideration of soil moisture problems are examined. A new theory is presented and briefly discussed. T. G. MORRIS.

**Soil conservation and erosion control measures on Reunion Island.** R. Guennelon (*Ann. Agron.*, 1955, **6**, 423—297).—The causes of erosion are discussed and measures for its control are described. A. H. CORNFIELD.

**Assessing the reliability of rainfall if monthly falls are not independent.** J. Glover, P. Robinson and J. Taylor (*J. agric. Sci.*, 1955, **46**, 387—388).—An amplification and correction of a previous paper (Glover and Robinson, *ibid.*, 1953, **42**, 275). A. G. POLLARD.

**Irrigation practices in Queensland.** A. Nagle (*Qd agric. J.*, 1954, **78**, 63—66, 125—132, 191—197 ; **79**, 1—14, 74—76, 125—129, 193—202).—Irrigation practices are described under the headings :—requirements for successful irrigation, methods of irrigation, instruments used for preparing the land, prep. of the land for border irrigation, special methods of water distribution, fundamentals of water application for surface irrigation, water requirements of irrigated crops, pumping water for irrigation. A. H. CORNFIELD.

**Sprinkler irrigation.** M. Velatta (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 312—348).—The history and development of sprinkler irrigation are outlined, together with uses and problems of the method, and its application to anti-parasite control. Various types of jets are discussed, and the results of sprinkler irrigation for various fruit crops are given, together with effects of atm. conditions. (84 references.) F. R. PAULSEN.

**Irrigation of clay soils.** A. Draghetti (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 295—311).—Some hydrophilous crops are adapted to heavy irrigation, but mesophilous or drought-resistant plants often suffer from the excessive water of a heavily irrigated clay soil, so that percolation through single or multiple small furrows is better in such cases. Water vol. should be adjusted to optimum soil moisture, not to water consumption of the plants. The max. capacity of the soil for water absorption is the basis for computing vol. of water for each soil and each irrigation. F. R. PAULSEN.

**Determination of amino-acids in soil hydrolysates by the Moore and Stein method.** F. J. Sowden (*Soil Sci.*, 1955, **80**, 181—188).—The non-basic amino-acids present in three soil hydrolysates were separated by column chromatography. The amino-acid pattern was similar in all three soils tested. The S containing acids, cystine and methionine were present in small amounts in all three hydrolysates. The percentage of amino-N accounted for was satisfactory. T. G. MORRIS.

**Reduction of nitrate by ferrous hydroxide under various conditions of alkalinity.** J. M. Bremner and K. Shaw (*Analyst*, 1955, **80**, 626—627).—Low results were obtained when $NO_3'$ was determined by reduction to $NH_3$ at room temp. with reduced Fe and $H_2SO_4$. Experiments suggested that some of the reduction occurs after the liquid has been made alkaline for distillation of $NH_3$, this residual reduction being effected by the $Fe(OH)_2$ then formed. They also showed that determination of $NH_3$ by distillation with MgO of solutions containing significant amounts of $NO_3'$ may give erroneously high results if appreciable amounts of $Fe^{\cdot\cdot}$ are present. In the conventional method for determination of $NH_3$ in soil by extraction with acid N-KCl solution and distilling the extract with MgO (Olsen; cf. Brit. Chem. Abstr., B., 1929, 951) the amount of $Fe^{\cdot\cdot}$ extracted is too small to cause interference by $NO_3'$. Distillation of 300-ml. solutions containing 1 mg. of $NO_3'$ and different amounts of $FeSO_4$ with MgO showed that no reduction of $NO_3'$ occurred when the concn. was 70 mg./l. and that quant. reduction occurred when it was 7 g./l. A. O. JONES.

**Loss of nitrogen as ammonia from water-logged paddy soil.** S. P. Gupta (*J. Indian Soc. Soil Sci.*, 1955, **3**, 29—32).—Laboratory tests simulating field conditions were carried out to determine the loss of $NH_3$ from $(NH_4)_2SO_4$ (60 lb. N per acre) applied as a top-dressing to a water-logged paddy soil (pH 8·4). Approx. 11% of the added N was lost as $NH_3$ during the first three days after application and 22% after 15 days. A. H. CORNFIELD.

**Determination of nitrate and ammonia in soil.** J. M. Bremner and K. Shaw (*J. agric. Sci.*, 1955, **46**, 320—328).—The method described involves extraction of soil with $K_2SO_4$–$H_2SO_4$ (to give pH 1·0—1·5 in the extract). The $NH_3$ content of the extract is determined by treatment with MgO in a modified Conway microdiffusion apparatus at 25°. In another portion of the extract $NH_3 + NO_3'$ is determined similarly after reduction of $NO_3'$ by $Ti_2(SO_4)_3$ added in advance of the MgO. A. G. POLLARD.

**Determination of the phosphorus status of soils.** G. Joret and J. Hebert (*Ann. Agron.*, 1955, **6**, 233—299).—Acid extractants were found to be unsatisfactory for determining the P status of calcareous soils. After an extensive study of the solvent effect of aq. $(NH_4)_2C_2O_4$ on $CaCO_3$, Ca phosphates, and phosphates adsorbed on clay, sesquioxides and humus, a method of assessing the P status of calcareous soils is proposed. Four g. of soil is shaken for 2 hr. with 100 ml. of 0·2N-$(NH_4)_2C_2O_4$. Volumetric and colorimetric methods for determining P in the extracts are described. Soils containing $P_2O_5$ >250 p.p.m. as extracted by this method generally made no response to application of P fertilisers. A. H. CORNFIELD.

**Determination of total organic phosphorus in soils.** W. M. H. Saunders and E. G. Williams (*J. Soil Sci.*, 1955, **6**, 254—267).—A study was made of two distinct methods of determining org. P in soils. In the ignition method org. P is the difference between inorg. P extracted with 0·2N-$H_2SO_4$ before and after ignition of the soil. In the extraction method org. P is extracted with 0·5N-aq. $NH_3$ or 0·1N-NaOH after acid pre-treatment of the soil. Results obtained by the extraction method varied with the type of acid pre-treatment. 0·1N-NaOH was a much better extractant of org. P than was 0·5N-aq. $NH_3$. The ignition method gave for most soils results very similar to those obtained by a double extraction with cold 0·1N-NaOH following pre-treatment with hot

0·1N-HCl. With some soils the ignition method gave somewhat higher results for org. P content. A. H. CORNFIELD.

**Mobilisation of phosphate in water-logged soils.** J. K. R. Gasser and C. Bloomfield (*J. Soil Sci.*, 1955, **6**, 219—232).—The effects of fermenting grass under anaërobic conditions on the mobilisation of Al and Fe phosphates, Ca phosphate, silicophosphate, and rock phosphate, basic $Fe^{\cdots}$ phosphate and $Fe^{\cdots}$ hydroxyphosphate, and natural and phosphated kaolin and montmorillonite were studied. Release of $PO_4'''$ occurred from both forms of Ca phosphates and from rock phosphate. There was no release of $PO_4'''$ or Al from Al phosphate, but both Fe and $PO_4'''$ were released from the $Fe^{\cdots}$ phosphates. Released $PO_4'''$ was re-adsorbed by basic $Fe^{\cdots}$ phosphate. The natural clays fixed $PO_4'''$ whilst the phosphated clays released $PO_4'''$. The results are discussed in relation to the mobilisation and movement of $PO_4'''$ in gleyed and artificially flooded soils. A. H. CORNFIELD.

**Relationship between pH and different forms of phosphorus in some soils of East Pakistan.** A. Karim and D. H. Khan (*Soil Sci.*, 1955, **80**, 229—233).—Samples of soil (26) from the same area with a pH range from 4·2 to 6·5 were analysed for org., adsorbed, sesquioxide-bound and total P. The org. P content increased with pH from 4·2 to 4·6, then decreased to a min. at pH 5·6, and increased again with increasing pH, although there was a wide scatter of points. Sesquioxide-bound P increased evenly with pH up to pH 5·3, then more sharply to a max. at pH 5·5 and then decreased. The amount of adsorbed P appeared to remain fairly constant until a pH of 5·0 was reached; a max. was recorded at pH 5·3 with a decrease with rise in pH above 5·3. The critical pH level of 5·3 to 5·6 is considered in the light of changes in the state of Fe and Al in the soil. T. G. MORRIS.

**Fertility studies on some New Brunswick soils. I. Soil phosphorus supply as shown by greenhouse and chemical tests.** A. A. MacLean, J. J. Doyle and F. G. Hamlyn (*Canad. J. agric. Sci.*, 1955, **35**, 388—396).—Applied P resulted in highly significant increases in yield and % P in ladino clover on all soils and in general the greater the P content of the crop grown without P fertiliser the smaller was the increase in yield from applied P. Soil P levels varied significantly between soil types and a positive correlation existed between these values and pH. E. G. BRICKELL.

**Changes in phosphorus compounds in soil and assimilation by plants.** A. Musierowicz (*Roczn. Nauk. rol.*, 1955, **70, A,** 557—581). —A critical review. The nature and sorption of org. and inorg. sources of P in soils are considered in relation to fertiliser action. A. G. POLLARD.

**Preparation and properties of some iron phosphates.** F. Scheffer and H.-G. Schulz (*Z. PflErnähr. Düng.*, 1955, **70**, 141—164).—The chemical and physical properties of some synthetic Fe and K–Fe phosphates are reported. Results are discussed in relation to phosphate fixation in soils. A. H. CORNFIELD.

**Improvement of phosphate availability in the laterite soils of the Nilgiris by the application of silico-phosphate.** A. Mariakulandai, S. Venkatachalam and T. R. Iyengar (*J. Indian Soc. Soil Sci.*, 1955, **3**, 15—22).—Application of silico-phosphate (rock phosphate, $Na_2CO_3$ and sand heated to 1300—1400° in presence of steam) to a laterite (pH 4·3—5·0) resulted in better growth of and P uptake by ragi (*Eleucine corocana*) in pot tests than did application of an equiv. amount of superphosphate. Application of $Na_2SiO_3$ (**I**) resulted in only slight growth improvement. Liming prior to applying the fertiliser improved the effect of superphosphate but not that of silico-phosphate or **I**. Green-manuring had little effect on the availability of P either in superphosphate or silico-phosphate. A. H. CORNFIELD.

**Effects of oxidants on solubility of soil phosphoric acid.** O. Colagrande (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 484—498).—Calcination decreases acid solubility of $P_2O_5$ in humus soils, increases it in the case of sandy, medium-and clayey soils, and has no effect on it in calcareous soils. Oxidising acids also increase solubility of $P_2O_5$ in humus, sandy, medium and clayey soils, but have no effect on that in chalky soils. These results are discussed. F. R. PAULSEN.

**Colorimetric determination of available potassium in soils.** J. Jacquet and Y. le Nir (*Ann. Agron.*, 1955, **6**, 499—505).—The soil is extracted with $CCl_3COOH$ (0·25—1·00%, depending on its $CaCO_3$ content) and the K is pptd. as $Co(NO_2)_6'''$. The ppt. is dissolved in HCl, the solution is evaporated to dryness and the colour developed after reaction with $NH_4CNS$ is measured absorptiometrically. A. H. CORNFIELD.

**Potassium availability from biotite, muscovite, greensand and microline as determined by growth of *Aspergillus niger*.** C. F. Eno and H. W. Reuzer (*Soil Sci.*, 1955, **80**, 199—209).—Quantities of the minerals, ground to pass an 80-mesh sieve, and further fractionated into particle sizes (i) 20—177, (ii) 2—20 and (iii) 0·2—2·0μ, were added to a nutrient solution inoculated with *A. niger*. After incubation at 28° for five days the K contents of the mycelium, culture solution and mineral residue were determined. With two exceptions, the wt. of mycelium increased with increasing K supply in the case of all the minerals in all sizes. The heaviest mycelia were produced by size (iii) biotite and muscovite; size (i) gave the lowest wt. With greensand all sizes produced almost the same wt. of mycelium for each K level. With microline, size (iii) produced the greatest wt. of mycelium. Growth of the fungus brought marked amounts of K into solution in the case of biotite, less in that of muscovite and very little from microline. Generally less K was released from the finer sizes than from the coarser in the case of greensand. In the cases of greensand and muscovite there was good correlation between the pH of the culture and the wt. of mycelium produced. T. G. MORRIS.

**Flame photometric determination of calcium in soil extracts.** K. Kropik (*Z. PflErnähr. Düng.*, 1955, **70**, 138—140).—Calcium is extracted from soil with $H_2CO_3$, and, after pptn. as $C_2O_4''$ and dissolution in HCl, is estimated with the flame photometer. Results obtained were very similar to those obtained by permanganometric determination of the $C_2O_4''$. A. H. CORNFIELD.

**Relation between soil degradation and manganese toxicity.** M. Ollaguier and P. Prevot (*Oléagineux*, 1955, **10**, 663—666).—The correlation between soil degradation, low pH and Mn toxicity in groundnut cultivation was confirmed. Application of $MnSO_4$ to a fertile soil reproduced the symptoms of Mn toxicity as observed in poor soils. The application of lime (2 ton/hectare) at sowing time reduced the symptoms of toxicity and, applied to pot cultivations, suppressed them altogether and substantially reduced the absorption of Mn. The period of lime application, its residual effect, and the influence of org. matter in soil were also studied. It is emphasised that Mn toxicity is found only in degraded soils and the best method of preventing it is the adoption of cultivation methods (rotation and green manure) which conserve the org. matter in the soil. J. S. C.

**Cobalt in meadow and pasture soils of certain highland regions.** A. Kabata (*Roczn. Nauk. rol.*, 1955, **70, A,** 609—615).—The Co content of the surface layer of the brown clays of the Carpathian area (8·2—15·5, average 14·3 p.p.m.) tends generally to increase, with the proportion of fine particles (mechanical analysis) present. A. G. POLLARD.

**Solubility of cobalt and its compounds in soil.** R. S. Young (*J. Soil Sci.*, 1955, **6**, 233—240).—The rate of leaching by rainwater of metallic Co and a no. of Co compounds (all finely powdered) added to the surface of columns of podsol soil, pH 6·0, was studied. Less than 0·2% of Co as metal was brought into solution in nine months. 1·2% of the Co as $CO_3''$ and 3·4% of that as $PO_4'''$ were released in the same period. 71—83% of the Co from Cl′, $NO_3'$ and $SO_4''$ were leached out of the soil. Residual soil Co was virtually completely sol. in 0·1N-HCl when the Co had been added as Cl′, $NO_3'$ or $SO_4''$. The residual Co from the $PO_4'''$ was also highly sol., whilst that from the $CO_3''$ and O″ were only slightly sol. in the 0·1N-HCl. A. H. CORNFIELD.

**Zinc in plants and soils.** T. J. Beckmann (*Qd agric. J.*, 1954, **79**, 24—25).—Zinc deficiency symptoms in plants and soils on which the element is likely to occur are described. Recommendations for curing the trouble under Queensland conditions are given. A. H. CORNFIELD.

**Variations of sulphur in soil under a continuous grain crop.** H. H. Mann (*J. Soil Sci.*, 1955, **6**, 241—247).—Changes in the S content of a freely-draining light sandy loam soil carrying spring barley and subjected to various manurial treatments are reported over 50 years. Over the period the total S level was not greatly different as between soil not receiving $SO_4''$ and that receiving fertilisers containing $SO_4''$. There was a gradual increase with time in the $SO_4''$ content of soils receiving $SO_4''$. There was a general decrease in total org. S over the 50 years; this decrease paralleled that of org. C. Humus-S (that peptised by $NH_4OH$) remained fairly const. over the period. S received by rain and other atmospheric sources was sufficient to maintain the S content of the soil at a reasonable level for crop growth. A. H. CORNFIELD.

**Electrometric determination of chloride in soil.** J. Benjaminsen and J. Jensen (*Tidsskr. Planteavl.*, 1955, **59**, 280—290).—The method involves the use of a Ag/AgCl electrode. Data obtained agree with direct determinations by Mohr's method. In an aq. medium the tendency for values in filtrates to exceed those in suspensions of soil is counterbalanced by use of 0·5N-$AgNO_3$ instead of water. A. G. POLLARD.

**Lime requirement of an acid sandy soil.** S. C. Mandal, S. K. Das and H. N. Mukherji (*J. Indian Soc. Soil Sci.*, 1955, **3**, 71—75).—The laboratory method of assessing lime requirement by noting the change in pH after adding varying amounts of $Ca(OH)_2$ agreed

closely with field changes arising from adding $CaCO_3$ to the soil. Rice yields were unaffected by increasing soil pH from 5·2 to 6·2. Wheat yields increased with pH up to about 6·2, but not with further increasing pH. A. H. CORNFIELD.

**Mobility ratios of clay membrane electrodes.** S. K. Bose (*J. Indian Soc. Soil Sci.*, 1955, **3**, 65—69).—The mobility ratios of the cation pairs Na/K, Na/Ca and K/Ca were determined using clay membrane electrodes. A. H. CORNFIELD.

**Soil humic acids. I. Chemical nature of humic nitrogen.** J. M. Bremner (*J. agric. Sci.*, 1955, **46**, 247—256).—Humic acid prep. isolated from mineral, fen and peat soils differed markedly in total N and N distribution after acid hydrolysis depending on whether 0·5M-NaOH or 0·1M-$Na_4P_2O_7$ (pH 7·0) were used for the initial extraction. The NaOH-extracted prep. had the higher N content and higher proportion of acid-sol. N and α-amino-N after hydrolysis with 6N-HCl; 20—60% of the total N present was not hydrolysed. Values of α-amino-N indicated that at least 31—48% of the N in the NaOH-extracted prep. and 20—35% of the N in the pyrophosphate-extracted prep. was in the form of protein; 3—10% of the N was present as amino-sugars. Paper chromatography of the acid hydrolysates showed that the humic acid prep. for the different soils did not differ markedly in amino-acid composition; 19 amino-acids, two unidentified ninhydrin-reacting substances, and amino-sugars were detected in every hydrolysate. A. H. CORNFIELD.

**Importance of organic matter in crop production.** G. W. Cooke and H. V. Garner (*J. roy. agric. Soc.*, 1954, **115**, 27—40).—Published experimental data relating to the value of org. matter, added to soil in the form of farmyard manure, straw, cereal stubbles, composts, green manures and leys, is discussed. The need is indicated for the assessment of the value of org. matter when used in presence of adequate supplies of essential plant nutrients. General recommendations are made for the use of org. manures on soils of different types. A. G. POLLARD.

**Rôle of organic matter in soil fertility.** N. R. Dhar (*Ann. Agron.*, 1955, **6**, 133—160).—Atm. N was fixed when org. materials (manure, straw, molasses, peat, etc.) were added to soils. The amount of N fixed was greater in light than in dark, and was particularly high in soils rich in Ca phosphates. The extent of fixation was similar in sterile and non-sterile soils. Direct addition of straw to soils resulted in greater fixation of N than did addition of composted straw. The extent of fixation of N due to org. matter additions is probably greater than that occurring during the growth of leguminous crops. Factors affecting losses of N are also discussed. A. H. CORNFIELD.

**Maintenance of organic matter in soils with straw and stable manure.** H. Kick and R. Dörr (*Z. PflErnähr. Düng.*, 1955, **70**, 124—137).—Application of straw + N to six different soils every second year over six years resulted in as satisfactory maintenance of the soil org. C level as did application of stable manure. The nature of the humus formed from both materials was very similar. Over the period the total N content of the soils decreased by an average of 6% where only mineral fertilisers were applied, but was virtually unchanged where either form of org. material had been applied. In the years of manure application cereal yields decreased in the order straw + N, mineral fertiliser, stable manure. In the years following the manure application yields decreased in the order mineral fertiliser, straw + N, stable manure. A. H. CORNFIELD.

**Colorimetric field test for organic matter in mineral soils.** S. N. Edson and R. H. Mills (*J. agric. Food Chem.*, 1955, **3**, 852—853).—A colorimetric procedure for determination of org. matter in mineral soils is described, based on determination of residual Cl, after shaking the soil sample with aq. $Cl_2$ for 1 min. and allowing to stand for 5 min. The suspension is filtered and to 2 ml. of the clear filtrate collected in a 16 × 150 mm. test tube, three drops of a prepared *o*-tolidine solution are added; the liquids are mixed by swirling and the intensity of yellow colour developed after standing for 15 min. is proportional to the amount of org. matter oxidised. The method is accurate to 0·5% and results of tests on 18 soil samples compare favourably with the standard chromic acid method. E. M. J.

**Use of sawdust in the soil.** F. Chippendale (*Qd agric. J.*, 1954, **78**, 199—200).—Some dangers associated with using sawdust as a soil mulch are discussed. If N is not applied with the sawdust, soil N may be rendered unavailable to plants during microbial decomposition of the straw. Some sawdusts contain essential oils which may have a deleterious effect on plant growth. Sawdusts from logs which have been treated with preservatives or pesticides (e.g., B compounds or pentachlorophenates) may contain sufficient of these materials to affect plant growth. A. H. CORNFIELD.

**Enzymes in soils. VI. Amylase.** E. Hofmann and G. Hoffmann (*Z. PflErnähr. Düng.*, 1955, **70**, 97—104).—Amylase activity in a no. of soils increased with pH up to about 5·5 and then decreased with further increase in pH. Optimum temp. for amylase activity was about 37°. Amylase activity in soils decreased with depth. In soils of pH 5·4—7·0 β-amylase was much more active than was α-amylase. Amylase activity was higher in soil under grass than in arable soil. A. H. CORNFIELD.

**Saccharase activity in soils.** E. Hofmann and K. Bräunlich (*Z. PflErnähr. Düng.*, 1955, **70**, 114—123).—Saccharase activity varied with soil type and increased with content of fine mineral particles (<0·01 mm.), humus and pH. Application of N, P and K increased saccharase activity, K in general having only a slight effect in this respect in comparison with N and P. Saccharase activity was somewhat higher in cropped than in fallow soils. A. H. CORNFIELD.

**Microbiology of acid soils. IV. Selected sites in northern England and southern Scotland.** J. G. Boswell (*New Phytologist*, 1955, **54**, 311—319).—Soil from nine localities was tested. With a few exceptions the soil pH was between 3·1 and 5·0 and the soil org. matter between 20 and 90% dry wt. *Penicillium* was the most abundant fungus (generally occurring in over 50% of the plates) followed by *Saccharomyces*, *Cephalosporium* and *Mucorales* (chiefly *Zygorrhincus*). *Pullularia*, *Cladosporium*, *Botrytis*, *Trichoderma*, *Verticillium* and *Stemphylium* were encountered much less frequently. L. G. G. WARNE.

**Influence of soil profile characteristics and nutrient concentrations on fungi and bacteria in Leon fine sand.** W. G. Blue, C. F. Eno and P. J. Westgate (*Soil Sci.*, 1955, **80**, 303—308).—In the groundwater podsol examined, org. matter decreased and pH increased with depth. Exchange capacity was high in the top layers and low in the $A_2$ horizon at 9—12 in. The K and Ca contents decreased with depth but that of P was relatively constant and high over the profile. The first two samples taken after fertilisation with a 5—5—8 mixture in varied amounts showed increases in the K, Na, P and $NO_3'$-N levels with increasing increments of fertiliser and at all depths. The third and later samples showed a rapid decrease in nutrient levels over the whole profile to the pre-fertilisation level. The no. of fungi and bacteria generally increased with increasing rates of fertilisation at all depths for the first two samples. The fungi decreased in no. with profile depth but the decrease was less at the higher fertilisation rates. Bacteria showed the same pattern but less distinctly. The no. of fungi and bacteria decreased sharply at the last two samples but larger no. of fungi remained in the fertilised than in the unfertilised soil; the reverse was true for bacteria. At the second sample the no. of organisms in the profile correlated very closely with the exchange capacity and the concn. of $NO_3'$-N and K. There was no correlation between soil Ca and no. of organisms. T. G. MORRIS.

**Ecology of fungi in soil.** D. Park (*Trans. Brit. mycol. Soc.*, 1955, **38**, 130—142).—Only native soil fungi effected deep penetration of plant remains in contact with the soil. Alien fungi, even if they colonised the material, remained superficial and aliens disappeared from such material if buried in the soil, whereas native fungi survived. L. G. G. WARNE.

**Reactions of *Aspergillus niger* to growth substances present in humus.** R. Chaminade (*Ann. Agron.*, 1955, **6**, 363—371).—Addition of composts or of aq. extracts of fresh and composted straw, or of $NH_4$ humate or humic acid extracted from straw accelerated the growth of *A. niger* in culture tests. Addition of compost ash had no effect on mould growth. A. H. CORNFIELD.

**Effect of B-vitamins and amino-acids on nitrification.** K. Gundersen (*Physiol. Plant.*, 1955, **8**, 136—141).—Nitrification by *Nitrosomonas* in pure culture or in association with certain heterotrophic soil bacteria was unaffected by thiamine, riboflavin, biotin, *p*-aminobenzoic acid, nicotinamide, pyridoxine, pantothenic acid or vitamin $B_{12}$ (separately or in combination) or by potato extract or garden soil. Tryptophan, glutamic acid and histidine (100 μg. per l.) were somewhat inhibitory and tyrosine and phenylalanine prevented nitrification completely. A. G. POLLARD.

**Metabolic processes in *Azotobacter chroöcoccum*, Beij.** F. Radler (*Arch. Mikrobiol.*, 1955, **22**, 335—367).—Relationships between some growth and environmental factors and the course of vital processes in *A. chroöcoccum* (dry matter production, increase in cell no., N fixation, carbohydrate consumption, $CO_2$ production, dehydrogenase and catalase activities) are examined. Growth and respiration of the organism were inhibited by 2 : 4-D: in non-aërated cultures 2 : 4-D was reduced to the aminophenol. A. G. POLLARD.

**Effects of low temperatures on nitrification of ammonia in soils.** O. E. Anderson and E. R. Purvis (*Soil Sci.*, 1955, **80**, 313—318).—Four soils were treated with aq. $NH_3$ or $(NH_4)_2SO_4$ (N equivalent 100 p.p.m.) and were incubated for periods of up to 42 days at varying temp. (2·8—11·1°). In an unlimed soil of pH 4·9, after 42 days

at 2·8° little more than traces of $NO_3'$ had accumulated. At 5·6° considerably more $NO_3'$ was formed in shorter time, aq. $NH_3$ being oxidised 2—3 times more rapidly than $(NH_4)_2SO_4$. After liming nitrification at 2·8° was little affected, but at higher temp. it began 2—3 weeks earlier, rates of nitrification being similar for the two sources of N. In three other soils (pH 5·9—6·3) there was some nitrification at 2·8° in six weeks but only in one soil was the amount appreciable. Differences in rate of nitrification due to soil type were maintained throughout the temp. range although there was a tendency for all soils to reach the same level of $NO_3'$ content after 42 days' incubation. T. G. MORRIS.

**Ammonification and nitrification in a strip mine spoil.** A. A. Wilson and G. Stewart (*West Virginia agric. Exp. Sta.*, 1955, *Tech. Bull.* 379, 15 pp.).—The ammonifying and nitrifying powers of non-vegetated and vegetated spoil samples from a coal strip-mined area were compared with those of samples from a normal soil nearby. In non-vegetated spoil (pH 3·29) asparagine was ammonified rapidly and urea slowly, whilst in the vegetated spoil and normal soil sample both materials were ammonified rapidly. Nitrification rate in the vegetated and unvegetated spoil was poor, probably because of low pH. Addition of $Ca(OH)_2$ improved nitrification. A. H. CORNFIELD.

**Effect of pesticides on nitrification in the soil.** H. A. Wilson (*West Virginia agric. Exp. Sta.*, 1954, *Tech. Bull.* 366, 14 pp.).—Incubation tests showed that soil nitrification was unaffected by treatment of the soil with five times the recommended annual rate of application in the case of Phygon, $C_6H_6Cl_6$, Systox, fixed Cu, parathion and chlordane. At 25 times the normal rate Phygon and $C_6H_6Cl_6$ inhibited nitrification. Ferbam inhibited nitrification even at the lower rate. Nitrification was temporarily inhibited by addition of ferbam (50 p.p.m.), ziram or zineb. The hemi-Co salt of dimethyldithiocarbamic acid had no inhibitory effect even at 1500 p.p.m. A. H. CORNFIELD.

**Effect of compounds of nitrogen on the nitrogen-fixing activity of nodule bacteria in soya-bean and pea ; relationships between them and the plants.** M. V. Fedorov (*Mikrobiologiya*, 1954, **23,** 534—543).—In sand cultures presence of excessive amounts of $NO_3'$ was associated with absence of nodules and of N fixation. With moderate proportions of $NO_3$ (50—60% of the normal amounts for sand cultures) the no. and size of nodules and the quantity of N fixed increased with rising $[NO_3']$. In water cultures existing nodules disappeared when the $NO_3'$ supply was raised to excessive levels but the nodule organisms remained in the roots and produced nodules again when the $NO_3'$ level was lowered. High $[NO_3']$ in contact with part of the root system did not affect nodulation and N-fixation in another part of the root in contact with moderate $[NO_3']$. Excessive N supplies to pure cultures of nodule organisms did not inhibit their activity. The action of high $[NO_3']$ on N fixation and nodule formation is probably due to physiological conditions within the plant. SOILS & FERT. (A. G. P.).

**Interrelationships between cellulose-decomposing bacteria and *Azotobacter*** Y. M. Voznyakovskaya (*Agrobiologiya*, 1954, No. 4, 81—85).—The two groups of organisms in soil and turf have a mutually stimulating action. SOILS & FERT. (A. G. P.).

**Chromatographic analysis of bacterial polysaccharides.** J. Dzulynska and E. Mikulaszek (*Acta biochim. polon.*, 1954/5, **1,** 191—196).—In hydrolysates of bacterial polysaccharides, glucosamine, galactose, (and/or glucose), mannose, xylose, arabinose and rhamnose occurred commonly. A fast-moving constituent, possibly a methyl-sugar, was present in some cases. In general bacterial polysaccharides contained xylose whereas those from actinomycetes contained arabinose. Mutation from rough to smooth variants was associated with loss of glucosamine, rhamnose and mannose from the polysaccharide hydrolysate. A. G. POLLARD.

**Antagonistic and stimulatory effects of soil micro-organisms upon *Sclerotium rolfsii*.** D. J. Morton and W. H. Stroube (*Phytopathology*, 1955, **45,** 417—420).—Of the soil-bacteria, -actinomycetes and -fungi (>1000 of each) examined, 0·2, 1·7 and 3·5%, respectively, inhibited the growth of *S. rolfsii* on nutrient media, and significantly reduced its virulence towards plants. Many of the organisms stimulate the growth of *S. rolfsii* on sucrose–nitrate–agar (without thiamine), but do not increase its virulence. P. S. ARUP.

**Gliotoxin in soils.** E. Evans and D. Gottlieb (*Soil Sci.*, 1955, **80,** 295—301).—The production of gliotoxin by two strains of *Penicillium terlikowski* and by *Trichoderma viride* in a black prairie loam has been examined. *Trichoderma viride* was the most productive. The toxicity was assayed by its effect on a suspension of spores of *Sclerotinia fructicola*. Gliotoxin accumulated in the inoculated sterilised soil but not in unsterilised soil. During the first 30 days of incubation the concn. of gliotoxin increased steadily in this soil to a max. after which it decreased again to an undetectable amount in 70 days. Four days after the inoculation the no. of fungal colonies in the soils decreased progressively. Added gliotoxin was capable of free existence in soils, but recovery by solvents was not complete and was not correlated with pH in the range 4·7—6·2. T. G. MORRIS.

**Effect of root secretions of pea and maize on the development of some soil micro-organisms grown in a plant rhizosphere solution.** N. V. Meshkov and R. N. Khodakova (*Microbiologiya*, 1954, **23,** 544—550).—Growth of *Azotobacter* was favoured more by root secretions of peas than by those of maize. *A. chroöcoccum* multiplied more than did *Rhizobium leguminosarum* in presence of maize-root secretions. Secretions from both species benefited non-sporing more than sporing bacteria. SOILS & FERT. (A. G. P.).

**Eighteen years of lysimeter studies. III.** E. M. Bastisse and S. Hénin (*Ann. Agron.*, 1955, **6,** 203—232).—Water and nutrient movement in lysimeter experiments over 18 years as affected by irrigation, plant cover and manurial application are examined and discussed. A. H. CORNFIELD.

**Soil tests for determining the fertiliser and lime needs of vegetable crops in the coastal plain soils of Virginia.** E. M. Dunton, jun., M. E. Taylor and R. B. Hall (*Virginia Truck Exp. Sta.*, 1955, *Bull.* 114, 22 pp.).—Details of the "rapid" soil testing methods used are described. A. H. CORNFIELD.

**Application of rapid chemical tests to diagnosis of mineral deficiencies in horticultural plants. III. Comparisons of tissue tests and total analyses for potassium, magnesium, calcium, phosphorus and nitrogen in potato and cauliflower.** D. J. D. Nicholas (*J. hort. Sci.*, 1955, **30,** 260—267 ; cf. *ibid.*, 1948, **24,** 3).—The results of rapid tissue tests for the above elements show satisfactory agreement with those of complete ash analyses, provided that the macerated material is extracted with acetate or citrate buffer solutions. Results obtained with the use of malonate or succinate buffers are less reliable. P. S. ARUP.

**Statistical techniques for inspection sampling.** M. J. R. Healy (*Trop. Agriculture, Trin.*, 1955, **32,** 10—19).—Mathematical. A. G. POLLARD.

**Effect of plot shape in reducing errors in tea experiments.** D. H. Laycock (*Trop. Agriculture, Trin.*, 1955, **32,** 107—114).—Data from two experimental sites are examined. On sloping land narrow plots running with the slope were preferable to broad plots running with the slope or very broad plots running across the slope. Effects of block shape were less consistent, but in two of three cases broad blocks were more satisfactory than narrow ones. A. G. POLLARD.

**Soil sampling errors and advisory analyses.** R. G. Hemingway (*J. agric. Sci.*, 1955, **46,** 1—8).—Sampling errors in pH and 1% citric acid-sol. P and K of 50 fields of differing soil type and manurial history are presented. Sampling errors were greater on soils which had received fertilisers and lime within three years of sampling than on those which had not. There was little difference in errors between wasteland and cultivated non-fertilised land, whilst grazed soils showed variations similar to those of fertilised soils. There was no general increase in error as the area which the samples represented increased. Errors of sampling, which were greater than analytical errors, were such that the common practice of classifying soils into six fertility groups was not justified. For routine purposes three groups should be sufficient. A. H. CORNFIELD.

**Deep ploughing on the farm.** A. Draghetti (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52,** Ser. ii, 271—276).—The history and importance of deep ploughing are outlined. Crop rotation allows max. use of soil org. matter, and a lucerne crop is best for virgin ploughed soil. The methods described lead to remarkable increase of forage crops and fertility. F. R. PAULSEN.

**Effects of tractor ploughing on the Black Soils of Malwa.** R. V. Tamhane and P. M. Tamboli (*J. Indian Soc. Soil Sci.*, 1955, **3,** 51—63).—Tractor ploughing of Black Cotton soils resulted in increased lower and upper plastic limits, plastic no. and sticky point of the soils in comparison with virgin soils. Aggregation was reduced by ploughing, in some cases by as much as 50%. Available nutrients, base exchange capacity and exchangeable bases were unaffected by tractor ploughing whilst org. C and total N were reduced. Ploughing generally slightly increased bacterial no. (total plate count) and nitrification and reduced the N-fixing capacity. A. H. CORNFIELD.

**Soils and fertilisers.** E. W. Russell (*J. roy. agric. Soc.*, 1954, **115,** 138—156).—Manurial trials on grassland for hay, recent investigations of the liming of acid soils and developments in the manufacture and use of P fertilisers are summarised. A. G. POLLARD.

**Residual effects of a phosphate fertiliser on a Wealden soil.** G. W. Cooke and J. K. R. Gasser (*J. Soil Sci.*, 1955, **6,** 248—253).—The residual effects, as measured by crop yields and soil analysis, of

superphosphate and poultry manure on a fine sandy silt loam, pH 5·6, are reported. A fresh dressing of superphosphate at 0·2 cwt. $P_2O_5$ per acre gave yields of potatoes similar to those obtained by applying superphosphate at 1 cwt. $P_2O_5$ per acre the year before. Poultry manure supplying 1 cwt. $P_2O_5$ per acre improved yields in both first and second years somewhat more than did superphosphate supplying the same amount of $P_2O_5$. The apparent recovery by crops of added P over six years ranged from 9 to 12%. The treatments had no significant effects of total, 0·5N-AcOH-sol., or 0·3N-HCl-sol. P in the cultivated layer, probably due to leaching of fertiliser P. A. H. CORNFIELD.

**Field experiments with Nitrophosphates.** A. H. Lewis (*J. agric. Sci.*, 1955, **46**, 287—291).—Three grades of Nitrophosphates containing water-sol. $P_2O_5$, respectively, 11, 16 and 33% of the total $P_2O_5$ were compared with superphosphate (90% water-sol.) as P sources for grass, swedes and potatoes. Nitrophosphate was slightly superior to superphosphate on grass ; the two root crops gave similar yields with both P fertilisers. A. G. POLLARD.

**Advantages of granulated superphosphate in soils of differing acidity.** M. L. Yankovich (*C. R. Acad. Agric. Fr.*, 1955, **41**, 528—529).—The results of Russian experiments are reviewed and compared with those obtained by the author, using granular and pulverised superphosphate on eight successive crops of wheat and millet in a clay–limestone soil (pH ~8). Granulation is beneficial in podsols and leached chernozems but has no advantage in clay–limestone soils. N. M. WALLER.

**Manurial value of serpentine and magnesium thermophosphates as compared with superphosphate in pot tests.** K. Boratyński, S. Roszykowa and Z. Turyna (*Roczn. Nauk. rol.*, 1955, **70, A**, 583—607).—The solubility in 2% citric acid of P in the thermophosphates is directly related to the fineness of grinding of the materials. Pot tests in sand and in soil confirm the relative availability of these fertilisers. A. G. POLLARD.

**Effect of composts and organo-mineral fertilisers on yields of spring wheat and sugar beet under irrigated conditions.** A. A. Lȳzin (*Agrobiologiya*, 1954, No. 4, 71—74).—The best method of utilising phosphorites in chernozems is to compost with dung using ≯10% of phosphorite. The compost (60 kg. per hectare) is best applied prior to autumn ploughing. SOILS & FERT. (A. G. P.).

**Liquid manure in the Seine maritime region.** E. Jouis and E. Hangard (*Ann. Agron.*, 1955, **6**, 301—309).—Data showing the density, total N and K, org. matter, org. N and water-sol. ash contents of pure and diluted urine, drainings from farmyard manure, and mixtures of urine and drainings from cows and pigs are presented. There was a close correlation between the N + K contents of the manures and both density and org. matter contents. Methods of conservation and utilisation of liquid manure are discussed. A. H. CORNFIELD.

**Effect of compost prepared with superphosphate on crop yields.** W. G. Walinjkar and C. N. Acharya (*J. Indian Soc. Soil Sci.*, 1955, **3**, 7—13).—Yields of marua and berseem in pot tests were increased to a greater extent by application of composted stable manure to which superphosphate had been added prior to composting than by application of equiv. amounts of superphosphate + manure (composted without addition of superphosphate). A. H. CORNFIELD.

**Use of a mechanical mixer in preparing fertiliser samples for analysis.** H. R. Allen (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 460—464).—The mixer holds two large containers for mixing the unground sample and two smaller ones for mixing portions after grinding. With samples containing <30 units of plant food, a one-pint sample could be obtained directly from the mixing chamber with a scoop. When higher-analysis mixtures contained ingredients varying greatly in particle size, it was necessary to spread out the sample and take portions from several places. A. A. ELDRIDGE.

## Plant Physiology, Nutrition and Biochemistry

**Photosynthesis.** C. P. Whittingham (*Endeavour*, 1955, **14**, 173—180).—Recent work on the mechanism and reaction steps of photosynthesis is reviewed, with particular reference to the fixation of $CO_2$, the rôle of chemosynthesis and the probable trends of future research. (16 references.) J. S. C.

**Light and mycorrhizal development.** K. F. Wenger (*Ecology*, 1955, **36**, 518—520).—In loblolly pines mycorrhizal development is most profuse in plants subjected to full natural light. L. G. G. WARNE.

**Photoperiodic cycles of lengths differing from 24 hours in relation to endogenous rhythms.** W. W. Schwabe (*Physiol. Plant.*, 1955, **8**, 263—277).—A repetition of Carr's experiments (cf. *ibid.*, 1952, **5**, 70) on the effects on the flowering of *Kalanchoë blossfeldiana* of light-breaks given at different times during the 60-hr. dark period of a 72-hr. cycle shows significant reductions in flowering when the breaks coincide with Bünning's three scotophile phases, although the responses for the second period are slight. In similar experiments with *Xanthium pennsylvaticum*, however, flowering occurred in all treatments without any response to the light-breaks. In other experiments with the above plants and another short-day plant (*Impatiens balsamica*) evidence was obtained of flowering in cycles differing widely from the 24-hr. cycle. Bünning's theory of the intervention of an endogenous rhythm in control of flowering is criticised in the light of the above findings. P. S. ARUP.

**Photoperiodism of *Kalanchoë blossfeldiana*. I. Effect of age on response to short-day treatment. II. Flowering of Dutch variety. Flowering of cuttings under short days.** A. F. Younis (*Physiol. Plant.*, 1955, **8**, 223, 229, 230—237).—I. Forty-five and (especially) the 66-week old plants show tardy or irregular responses to photoperiodic induction. From a review of the work of Harder *et al.*, it appears that max. responsiveness occurs at 26 weeks of age.

II. The Dutch variety of the plant requires short-day conditions for flowering, the max. permissible day-length being 11·5 hr. Cuttings of the common variety show tardy responses to photoperiodic induction, especially when the application of the stimulus is delayed until 2—3 weeks after planting. The delay is probably due to a deficiency in nutrients, and/or of water contents of the plant. P. S. ARUP.

**Short-time fixation of labelled carbon dioxide by barley leaves under steady state conditions.** A. R. Krall (*Plant Physiol.*, 1955, **30**, 269—271).—A device is described which will expose barley leaves to $^{14}CO_2$ for times as short as 0·10 ± 0·03 sec. before inactivating the tissue rapidly in boiling water and ethanol. The rate of fixation remained essentially constant over a range of 0·10 to 154 sec. E. G. BRICKELL.

**Carbon dioxide fixation and ion absorption in barley roots.** L. Jacobson (*Plant Physiol.*, 1955, **30**, 264—269).—Young excised barley roots fixed from 1·18 to 7·02% of $^{14}C$-labelled $CO_2$ in 3 hr. and in all cases malate formation accounted for a large portion of the $^{14}C$ present in the tissues, the malate being preferentially and approx. equally labelled in the α- and β-carboxyl positions. Glutamine, labelled in the α-carboxyl position, which appeared in the analysis as pyrrolidonecarboxylic acid, contained much less $^{14}C$ than the malate, but considerably more than any other identified constituent. E. G. BRICKELL.

**Identification of the carbon dioxide burst in *Chlorella* using the recording mass spectrometer.** A. H. Brown and C. P. Whittingham (*Plant Physiol.*, 1955, **30**, 231—237).—The observations of Emerson and Lewis (*Amer. J. Bot.*, 1941, **28**, 789—804) were confirmed. E. G. BRICKELL.

**Persistent rhythms of $O_2$ consumption in potatoes, carrots and the seaweed, *Fucus*.** F. A. Brown, jun., R. O. Freeland and C. L. Ralph (*Plant Physiol.*, 1955, **30**, 280—292).—The mean daily rate of respiration of the potato showed a 15-day cycle with max. near the times of full moon and new moon ; carrots showed a min. about the time of full moon and max. at about the third quarter and new moon ; *Fucus* displayed cyclic variations with max. at 6- and 7-day intervals. The average daily pattern for potatoes was found to show a max. at 4 to 5 a.m. and lesser max. at 10 a.m., 3 p.m. and 10 p.m. ; carrots gave a conspicuous max. at 4 to 5 a.m. and a min. just after noon ; *Fucus* exhibited max. at 3 to 4 a.m. and 1, 6 and 11 p.m. with principal min. at 7 a.m. and 8 p.m. A significant correlation was also noted between the hourly rates of respiration and the concurrent rate of change in barometric pressure for all three plants. E. G. BRICKELL.

**Growth studies in woody species. VII. Photoperiodic control of germination in *Betula pubescens*, Ehrh.** M. Black and P. F. Wareing (*Physiol. Plant.*, 1955, **8**, 300—316 ; cf. *ibid.*, 1953, **6**, 692 ; 1954, **7**, 157).—Light requirements for germination are shown by the unchilled seeds of the tree, but not by seeds which have been chilled at 1—5° during four weeks. At 15°, the % germination of the unchilled seeds is increased in proportion to the length of the daily period of illumination, but at 20° germination is independent of photoperiodic conditions, and shows max. results after a single exposure to light. The stimulating effect of a single exposure to red light can be neutralised by i.r. irradiation. An inhibitory influence on germination (which is overcome by exposure to light) is probably located in the pericarp. P. S. ARUP.

**Carbon dioxide as carbon source and narcotic in photosynthesis and growth of *Chlorella pyrenoidosa*.** E. Steemann Nielsen (*Physiol. Plant.*, 1955, **8**, 317—335).—With the use of appropriately low concn. of the alga in aërated carbonate–bicarbonate buffer solutions, and with an improved method of aëration through sintered glass, it is shown by five different methods that max. rates of photosynthesis are attained

at 0·01—0·03% (by vol.) of $CO_2$. At high concn. of $CO_2$ (>1%) and under full illumination, the rates of photosynthesis decrease. At low pH (4·7) or under deficient illumination, the rates may increase with increasing concn. of $CO_2$ up to ~3%, beyond which they decline. The implications of these findings are discussed with reference to results obtained by other investigators. (22 references.)
P. S. ARUP.

**Conversion of protochlorophyll into chlorophyll-a in continuous and intermittent light.** H. I. Virgin (*Physiol. Plant.*, 1955, **8**, 389—403).—The conversion in intact etiolated barley leaves or in suspensions of the ground leaves in glycerol can be observed accurately by a spectrofluorimetric method, which, in comparison with the spectrophotometric method, allows of the use of much smaller amounts of material, and dispenses with the necessity of isolating and purifying the pigments. The bimol. order of the conversion is confirmed. Intermittent illumination (with a constant light period of 3·3 and dark periods of 13·2—63·3 millisec.) causes, in comparison with continuous illumination, no change in the conversion yields at 0°. On the reasoning of Emerson and Arnold (cf. *J. gen. Physiol.*, 1932, **15**, 391), it follows either that the conversion includes no non-photochemical reactions, or that the half life time of the intermediates is very short. P. S. ARUP.

**Comparison between etiolation and pigmentation of chlorophyll-deficient and normal shoots of *Tradescantia albiflora* var. *albovittata*.** K. Drumm (*Planta*, 1955, **46**, 92—112).—The blue and the orange are the most effective ranges of the visible spectrum for remedying internodal elongation and leaf deformation due to insufficient illumination. The qual. composition of the carotenoids of leaves containing ~10% of the normal amount of chlorophyll is similar to that found in normal leaves. Chlorophyll contents of the former leaves (~1% of the normal) show an abnormal ratio between chlorophyll-*a* and -*b*, viz. 1·56 : 1. The bleached leaves contain a pigment belonging to the phorbine group (probably methyl-pyrophæophorbide) which shows absorption max. at 410, 505, 535, 610 and 668 mμ. The same pigment occurs in chlorophyll-deficient leaves of *Xanthosoma maximiliani*. Root extracts contain no such pigment; the phorbin pigment is probably connected with the biosynthesis of chlorophyll. (33 references.) P. S. ARUP.

**Metabolism of chlorogenic acid and in higher plants.** H. Ruckenbrod (*Planta*, 1955, **46**, 19—45).—The author's method for the determination of chlorogenic acid (**I**) in plant material (cf. *Ber. dtsch. bot. Ges.*, 1955, **66**, 75) is described. Contents of **I** in sunflower seeds increase during ripening and decrease during germination; respiration and fat contents increase during ripening. The photosynthesis of **I** and anthocyanin in the leaves of *Hedera helix* is greatly promoted by nutrition with aq. sucrose, but not by nutrition with aq. mannitol, inositol or tyrosine; this synthesis is inhibited by NaF or 2 : 4-dinitrophenol, but not by Na fluoroacetate or by moderate $CHCl_3$-narcosis. Fluoroacetate, however, inhibits anthocyanin synthesis. A substance resembling **I** (probably an isomer) is found in the ivy leaves. (54 references.) P. S. ARUP.

**Photosynthesis and respiration of three blue-green algæ.** W. A. Kratz and J. Myers (*Plant Physiol.*, 1955, **30**, 275—280).—*Anabæna variabilis*, *Anacystis nidulans* and *Nostoc muscorum G* were studied by conventional Warburg manometry. In terms of their light intensity curves they showed no feature markedly different from those of *Chlorella*. E. G. BRICKELL.

**Respiration of barley plants. VIII. Nitrogen assimilation and respiration of the root system.** A. J. Willis and E. W. Yemm (*New Phytologist*, 1955, **54**, 163—181).—Excised roots of barley fed with $NH_4$ salts have their respiration rate increased. This effect is greatest with N-deficient roots, rich in carbohydrate.
L. G. G. WARNE.

**Connexion between respiratory gradient and growth rate in wheat roots.** L. Eliasson (*Physiol. Plant.*, 1955, **8**, 374—388).—A method for germinating and culturing wheat under sterile conditions is described. Respiration rates of the different parts of the seedling roots suspended in a solution containing inorg. salts and glucose are determined, with or without additions of indolylacetic acid (in inhibitory amounts) or of α-3-indolyl*iso*butyric acid. Respiration rates in the extension zone reach the same (approx.) level, irrespectively of growth rates or of the growth-substances added, but the duration of the period of increased respiration depends on the period of growth. The period and extent of growth and elongation (as compared with those of the controls) are primarily determined by the added growth substances, and appear in their turn, to constitute the factor which regulates increases in dry and fresh wt. and total N content. The direct connexion between increases in respiration and N content is confirmed. The connexion between root-elongation and the various metabolic processes is discussed. (37 references.)
P. S. ARUP.

**Passive components in the ion absorption of the plant. I. Zonal ion and water absorption in Brouwer's experiments.** B. Hylmö (*Physiol. Plant.*, 1955, **8**, 433—449).—Brouwer's theory of ion uptake is re-examined. The uptake by broad bean roots is prevented by inhibitory substances, e.g. KCN or 2 : 4-D: the remaining "passive" uptake is directly related to the water intake. The water flow into plants may be restricted by addition of sugar to the external medium without a corresponding reduction in ion intake. The sugar addition may increase the ion intake of deficient plants: it did not affect the relation between waterflow and the passive intake of ions. The Hagen–Poiseuille law of ultrafiltration is applicable to plant roots.
A. G. POLLARD.

**Specificity of voltage potentials in cation uptake by plants.** E. L. Breazeale and W. T. McGeorge (*Soil Sci.*, 1955, **80**, 319—324).—Single tomato plants were grown in solutions of various nitrates with the negative lead of the electropode attached to the plant and the positive lead dipping into the salt solution. The potential applied varied with the cation under test, e.g. 2·10, 2·13, 2·20 or 2·23 volts for Na, K, Ca and Mg respectively, but the current was maintained constant at 10 microamps, for seven days. The plants were then removed and the residual cation concn. was determined. The greatest uptake of each cation occurred when the potential was at a specific level for the ion. Uptakes increased with increasing salt concn. When the plants were grown in mixed solutions of equal concn. the same specificity of cation potential was evident. When the salt solutions were of unequal cation concn. there was an apparent substitution of one cation for another. T. G. MORRIS.

**Kinetics of exchange between adsorbents. IV. Unequal valency ion pairs.** C. Krishnamoorthy and A. D. Desai (*Soil Sci.*, 1955, **80**, 325—333).—The equilibrium in an adsorbent system for unequal valency ion pairs may take up to six months to be established. Given time, the observed and calculated equilibrium constants agree well. The amount of ion exchanged between unequal valency pairs is proportional to $t^{\frac{1}{2}}$, where $t$ is time, and the fractional equilibrium attained in a given time is independent of the initial concn. Gel diffusion is characteristic of exchange of unequal valency ion pairs. It is suggested that the diffusion constant for exchange between two adsorbents will vary inversely as the square of the radius of the larger particles only. T. G. MORRIS.

**Penetration of spray solution into leaves of higher plants.** I. Shetlik and A. Trukova (*Czekhoslov. Biol.*, 1954, **3**, 237—239).—A few leaves were sprayed with aq. $NaH_2PO_4$ labelled with $^{32}P$ or with aq. $Na_2SO_4$ containing $^{35}S$. After three days all unsprayed parts of the plants showed radioactivity. The salts penetrated through stomata and also by other routes.
SOILS & FERT. (A. G. P.).

**Transport of water in wood.** H. Lundegårdh (*Ark. Bot.*, 1955, **3**, 89—119).—The mobile water in tree trunks, i.e. that constituting the transpiration stream, exists in continuous threads mainly in the medium-sized tracheids of diameter about 10μ. Wider tracheids contain much air. In very narrow tracheids the capillary resistance to water movement is very high. Detailed consideration is given to the physical condition of water throughout the tree stem. Supporting experimental data include sap analyses, water distribution in tissues, size distribution of tracheids and water transport rates.
A. G. POLLARD.

**Relative distribution of potassium and rubidium-86 within maize plants grown in the field.** W. Z. Mackie and M. Fried (*Soil Sci.*, 1955, **80**, 309—312).—Maize was grown in the field with three levels of applied K. The centre row of plants on each plot received $K_2O$ tagged with $^{86}Rb$. The Rb–K ratio decreased with time indicating that K was absorbed by the roots from beyond the fertilisation zone. The ratio on the control plots was higher than that on the fertilised plots. The ratio was higher in the plants fertilised with 80 lb. of $K_2O$ than in those receiving 20 lb., although the ratio was the same in both fertilisers. The ratio in the tassels was significantly higher than in the leaves, nodes and internodes at all levels of K treatment. Similarly it was higher in the ears than in the non-reproductive part of the plant at the 80-lb. treatment level. T. G. MORRIS.

**Distribution of potassium, rubidium, cæsium, calcium and strontium within plants grown in nutrient solutions.** R. G. Menzel and W. R. Heald (*Soil Sci.*, 1955, **80**, 287—293).—Millet, oats, buckwheat, sweet clover and sunflower were grown in nutrient solutions containing adequate supplies of nutrients and $^{86}Rb$, $^{134}Cs$ or $^{89}Sr$. In plants harvested soon after flowering, data for the "distribution factor", viz. the ratio of bases in the plant parts divided by the same ratio in the nutrient, was determined. In all whole plants the distribution factor for Rb–K averaged 0·84, i.e., K was concentrated relative to Rb in the plant. Similarly the Cs–K factor showed greater discrimination against Cs than against Rb. The factor Sr–Ca showed that more Ca than Sr was absorbed. The factor for each pair was significantly different from unity, but the differences

between species for the same pair of elements were not significant. In millet and oats the Rb–K factor was higher for the roots than for the aërial portions. With buckwheat the factors were higher in the leaves and flowers than in the petioles, stems and roots. In sweet clover the factor was higher for leaves and roots than for stems and in sunflower values for flowers, upper leaves and petioles and roots were higher than for the rest of the plant. The Cs–K factor varied similarly to that of Rb–K except that it was relatively lower in the young parts of the plants. Sr was concentrated in the roots and the Sr–Ca ratio became progressively lower in the stems, petioles and leaves. T. G. MORRIS.

**Specific effects of adsorbed ions on plant growth. I. Effect of different combinations of calcium, magnesium and sodium on barley seedlings.** M. M. Elgabaly (*Soil Sci.*, 1955, **80,** 235—248).—Na-, Ca- and Mg-saturated Amberline IR-100H resins were used in sand cultures to provide varying proportions of bases in the three cation pairs. Barley seeds were germinated and then grown in the cultures for five weeks, harvested and analysed. With all systems, the growth as measured by the yield was better with mixed cations than with single cations. Max. yield occurred with 80Ca/20Na, 80Mg/20Na and 20Ca/80Mg. Length of shoots and roots were also max. at or near these concn. ; roots were more sensitive than shoots to the environment. In both Na systems, leaf burn occurred when the % of Na approached 100. In the Na/Ca system, as the supply of Na increased, the Na content of the barley increased to a max. at 60% Na saturation, and then fell slightly at 100% of Na. The Mg and K content of the barley decreased with increasing Na supply and with 100% of Na the plants contained practically no K. The Ca content of the plants was unaffected by the level of Na supply. The Na/Mg and Na/Ca systems produced parallel results. With increase in Mg level in the Ca/Mg system the Mg and Ca contents of the plants fell but the K was unaffected. Within limited ranges of concn. any one adsorbed cation depressed, but any pair of cations stimulated the growth of the barley compared with that in pure sand. Ca with Na stimulated growth over a wider range than did Mg with Na. Ca and Mg together had a wider range still. T. G. MORRIS.

**Calcium–magnesium nutrition with special reference to serpentine soils.** R. B. Walker, H. M. Walker and P. R. Ashworth (*Plant Physiol.*, 1955, **30,** 214—221).—Tomato, common sunflower, buckwheat and three annual species native to California serpentine areas were grown in pot cultures in serpentine soils in which the exchangeable Ca and Mg contents were artificially varied. Sunflower and buckwheat were also grown in culture solutions with varying Ca/Mg ratios. Yields of the crop plants were markedly depressed in soils having 20% or less of exchangeable Ca and made little or no growth with $<10\%$. The native species were unaffected by exchangeable Ca levels between 6 and 82% and growth was not drastically reduced (10—20%) in the range 3—5%. E. G. BRICKELL.

**Effect of magnesium fertilisers on seed quality.** M. M. Mazalva (*Agrobiologiya*, 1954, No. 4, 125—129).—In pot trials with millet, water-melon and maize Mg increased yields, improved seed quality and, in water-melon, raised the sugar content.
SOILS & FERT. (A. G. P.).

**Minor elements in soils.** D. F. Stenuit (*Agricultura*, 1955, **3,** 129—155).—A review. The rôle of trace elements in plant nutrition, deficiency symptoms, relevant soil conditions and analysis, correlation of deficiencies and the relation of these elements to animal health are discussed. A. G. POLLARD.

**Relationship between nickel toxicity and iron supply.** W. M. Crooke (*Ann. appl. Biol.*, 1955, **43,** 465—476).—For a fixed Fe supply absorption of Ni by oat plants in culture tests increased with pH (4 to 7). Ni uptake and necrotic and chlorotic symptoms were reduced with high Fe in the nutrient solution. Absorption of Ni and intensity of necrosis increased with increasing Ni–Fe ratio in the nutrient solution. With solutions having constant Ni–Fe ratio toxicity symptoms increased with the absolute amount of Ni supplied. Ni reduced the Fe content of roots and tops. In Ni-toxic plants the Mg, Ca and P contents of the tops and the K, Ca and P contents of the roots were higher than in healthy plants, whilst the K content of the tops and the Mg content of the roots were lower. Similar results were found with tomato. A. H. CORNFIELD.

**Relationship between nickel-toxicity symptoms and the absorption of iron and nickel by oats.** W. M. Crooke and A. H. Knight (*Ann. appl. Biol.*, 1955, **43,** 454—464).—Over the 70-day period from germination to maturity, the Fe content of oat plants (grown in culture) that showed symptoms of Ni toxicity changed little, but the Ni content increased rapidly for about 30 days and then decreased slowly. Chlorosis increased over 40 days and then decreased. Chlorotic areas of the leaf had a low, whilst necrotic areas had a very low, Fe content in comparison with that of healthy areas. Necrotic areas of leaves of oats grown on high-Ni soil had a lower Fe content than had healthy leaves. A. H. CORNFIELD.

**Chelates as correctives for chlorosis.** R. S. Holmes and J. C. Brown (*Soil Sci.*, 1955, **80,** 167—179).—Soya-beans were grown on a calcareous silty clay loam of pH 7·9 known to induce chlorosis. Five chelates EDTA, HEEDTA (*N*-hydroxyethyl-ethylenediaminetetra-acetic acid), DTPA (diethylenetriaminepenta-acetic acid), CDTA (*cyclo*hexanediaminetetra-acetic acid) and APCA (an aromatic polyamino-carboxylic acid) were added to the soil at different but comparable rates. Plants were harvested as soon as chlorosis developed in the lowest chelate treatment. EDTA, HEEDTA and CDTA did not cure chlorosis, but DTPA ($\ll$120 p.p.m.) did so. APCA was successful at all levels ; 3 p.p.m. partly corrected the condition and 10 p.p.m. prevented chlorosis completely. APCA diminished the uptake of Mn and Cu by the plants. P and K levels were higher in plants on the control plots than in those given APCA at levels of $\ll$660 p.p.m. Yields increased with increasing levels of APCA. With increasing levels of CDTA from 0 to 480 p.p.m. yields decreased and chlorosis appeared, at all levels. Fe contents of the plants increased with increasing DTPA levels, but P, Ca and K levels tended to be lower in treated than in untreated plants. CDTA–Fe added to the soil cured chlorosis. The effectiveness of DTPA–Fe in curing chlorosis decreased in successive crops. Use of radio-iron showed that DTPA released fixed Fe from the soil and made it available to plants. DTPA tagged with $^{14}C$ was absorbed by the plants and distributed throughout the aërial portions. DTPA sprayed on to chlorotic leaves did not correct the condition.
T. G. MORRIS.

**Morphogenesis of leaves. X. Relation between nitrogen nutrition, rate of respiration and rate of ageing in fronds of *Lemna minor*.** E. Wangermann and H. S. Lacey (*New Phytologist*, 1955, **54,** 182—198).—In culture, max. frond length of life and min. rate of ageing occurred at low N levels. Fronds of low-N plants had a lower respiration rate than those from high-N plants. Fronds periodically kept in N gas to inhibit respiration had their life extended.
L. G. G. WARNE.

**Ionic species in orthophosphate absorption by barley roots.** C. E. Hagan and H. T. Hopkins (*Plant Physiol.*, 1955, **30,** 193—199).—*Hordeum vulgare*, var. Atlas 46 was studied. In excised roots $H_2PO_4'$ is absorbed through one site on the root and $HPO_4''$ through another. OH′ competitively inhibits absorption of both $H_2PO_4'$ and $HPO_4''$. Neither $[H^+]$ nor $[OH']$ in the external medium affects the concn. of P at either effective absorption site. Mechanism of breakdown of both kinds of intermediate phosphate compounds formed at the absorption sites involves cleavage of an R–O bond.
E. G. BRICKELL.

**Phosphorus–iron relationship in genetical chlorosis.** P. C. DeKock and A. Hall (*Plant Physiol.*, 1955, **30,** 293—295).—The P/Fe ratio is higher and the Ca/K ratio is lower in chlorotic than in normal green leaves. E. G. BRICKELL.

**Uptake of phosphate by excised mycorrhizal roots of beech. VII. Active transport of $^{32}P$ from fungus to host during uptake of phosphorus from solution.** J. L. Harley and J. K. Brierley (*New Phytologist*, 1955, **54,** 296—301).—Active transport from fungus to root is reduced when the roots are washed in aq. $PO_4'''$. Rapid transport is resumed when the roots are replaced in a $PO_4'''$-free medium.
L. G. G. WARNE.

**Effect of light intensity on the phosphorus metabolism of spring wheat at different periods.** V. V. Rachinskiĭ, B. E. Kravtsova and E. I. Knyazyatova (*Biokhimiya*, 1954, **19,** 513—520).—Light intensity did not affect the uptake of $^{32}P$ by roots nor the distribution of P compounds in various parts of the plant. During shooting and ear formation P metabolism increased with light intensity.
SOILS & FERT. (A. G. P.).

**Continuous culture of excised rye roots.** E. H. Roberts and H. E. Street (*Physiol. Plant.*, 1955, **8,** 238—262).—The continued cultivation, without diminution in growth-rate, of the excised roots in a modified White's medium is rendered possible by additions either of Difco yeast-extract or of D- or L-tryptophan. Growth is also stimulated by B.D.H. peptone. Additions of nicotinic acid, L-kynurenine, gramine or skatole are ineffective, but indol-2-ylacetic acid (**I**) or indol-2-ylacetonitrile (**II**) is as effective as D- or L-tryptophan at mol. concn. of 0·0025% of the optimum mol. concn. for tryptophan. The activity of tryptophan depends on its modification during sterilisation (or refluxing in water). Evidence based on observations of the partition of the " activity " between $Et_2O$ and $H_2O$ and on chromatographic investigations does not support the view that the " activation " is due to the formation of **I** or **II**. The culture technique is successful with 10% only, of the rye grains tested, and is not applicable to other cereal grains. (56 references.)
P. S. ARUP.

**Influence of organic acids on respiration, ammonium-uptake, and free amino-acids of *Chlorella*.** O. Kandler and H. Ernst (*Planta*, 1955, **46,** 46—69).—Among the org. acids of physiological importance which are examined for stimulating effects on the respiration of the

impoverished alga, acetic and pyruvic acids only have effects comparable with those of glucose. The effects of the other acids are, by comparison, weak and transitory. The effects of org. acids (even those which promote respiration) on $NH_4$-uptake are negligible in comparison with those of glucose. The effects of org. acids on the composition of the free amino-acids of the alga are illustrated by chromatograms; they include the rapid decarboxylation of glutamic acid by simple org. acids. (35 references.) P. S. ARUP.

**Comparative physiology of green and albino maize seedlings.** H. Seltmann (*Plant Physiol.*, 1955, **30**, 258—263).—Albino maize seedlings have higher sol. N and lower protein-N contents than have green seedlings; $O_2$ uptake per g. of dry wt. was also 20 to 30% higher. Probably the metabolism of the light-grown albino maize seedlings is primarily non-phosphorylative. E. G. BRICKELL.

**Development of bean seeds (*Phaseolus vulgaris*, L.).** J. R. Loewenberg (*Plant Physiol.*, 1955, **30**, 244—250).—Development is described per cell and per organ in terms of dry wt., N content, P content, fresh wt. and rate of $O_2$ uptake. Comparison of the per-cell value of various plants indicates that while the cells differ in size and content, their rates of $O_2$ uptake are comparable. E. G. BRICKELL.

**Bacteria in higher plants.** S. Tonzig and L. B. Orsenigo (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 97—105).—Bacterial cultures have been prepared from leaves, roots and other organs of many non-leguminous plants of higher orders, of a no. of families, as these bacteria appear to differ from those of root-nodules of leguminosæ. F. R. PAULSEN.

**Fat metabolism in higher plants. VI. Incorporation of $^{32}P$ into peanut mitochondrial phospholipins.** M. Mazelis and P. K. Stumpf (*Plant Physiol.*, 1955, **30**, 237—243).—The process is described, an adenine nucleotide, $Mg^{++}$, and a Krebs cycle intermediate being required. The phospholipin which becomes labelled is unknown but chromatographic studies show that it is not phosphatidylcholine, phosphatidylserine, phosphatidylethanolamine, or a phosphatidic acid. E. G. BRICKELL.

**Atomic energy in agriculture.** K. L. Englund (*J. agric. Food Chem.*, 1955, **3**, 826—831).—The use of radioactive materials in agriculture is reviewed, covering radiation effects and applications, radio-isotopes, tagged agricultural chemicals, photosynthesis, the value of radioisotopes, e.g., in the study of enzyme systems in fungi, bacteria, insects and plants, as useful laboratory tools, and indications of future applications and developments. E. M. J.

**Radiation sensitivity of dormant and germinating barley seeds.** C. F. Konzak (*Science*, 1955, **122**, 197).—Dry and wet (soaked for 24 hr.) barley seeds were exposed to either *X*-rays or to neutron irradiation, and then sown. The water content of the seeds was determined at the time of irradiation and the seedling height after 14 days was taken as a measure of injury. Moisture content played an important part in the response to irradiation, especially in the case of *X*-rays, the toxic effect of which increased with increasing moisture content. Thermal neutrons had a toxic effect which did not vary greatly with moisture content. The mode of action is discussed. T. G. MORRIS.

**Influence of the moisture content of seeds on their sensitivity to *X*-rays.** M. Lefort and L. Ehrenberg (*Ark. Bot.*, 1955, **3**, 121—124).—The sensitivity of tomato seeds to *X*-ray injury, assessed by length of hypocotl and length and breadth of cotyledons, increased with the R.H. with which the seeds were in equilibrium. A similar relation occurs with barley seed. A. G. POLLARD.

**Effects of ultraviolet radiation and calcium and their interaction on salt absorption by excised mung bean roots.** T. Tanada (*Plant Physiol.*, 1955, **30**, 221—225).—The effects of Ca, u.v. radiation, dil. aq. HCl and NaCl on salt absorption by excised roots of *Phaseolus aureus* has been investigated using $^{86}Rb$ and $^{32}P$ as tracers. Ca enhanced the take up of both Rb and P, the locus of action being in or near the outer surface of the cytoplasm. Dil. HCl and NaCl increased Rb uptake but decreased P absorption, the effects being reversed by Ca. Irradiation (2537 A.) increased Rb absorption, except in the presence of Ca, and decreased P absorption to a slight degree though more so in the presence of Ca. E. G. BRICKELL.

**Mechanism of phytohormone damage by ionising radiation. I. Radiosensitivity of indolylacetic acid (IAA).** S. A. Gordon and R. P. Weber (*Plant Physiol.*, 1955, **30**, 200—210).—No unusual lability of the auxin exposed *in vitro* to *X*- and $\gamma$-radiation was observed on varying the auxin concn. or purity, the $O_2$ concn. or $H^+$ concn. of the solution, radiation energy, dose rate, or method of assay. Ionic yields were near unity. Acid auxin extracted from the plant exhibits an intrinsic radiosensitivity not significantly higher than synthetic IAA. E. G. BRICKELL.

**Growth substances and plant development.** L. J. Audus (*Endeavour*, 1955, **14**, 205—211).—The pattern of growth substances is studied by following the cycle of development of the flowering plant from its inception at the fusion of male and female nuclei in the flower, to maturity and seed-formation. (15 references.) J. S. C.

**Separation of acidic and non-acidic growth-substances.** P. Larsen (*Physiol. Plant.*, 1955, **8**, 343—357).—In separations based on partition between $Et_2O$ and $H_2O$, the use of tartaric acid causes the formation of a substance which inhibits the action of indolylacetic acid (**I**), and which passes into the $Et_2O$-phase. Tartaric acid can be used for acidifying the aq. phase when the amount of **I** present is $\leq 0{\cdot}08$ $\mu$g., but with smaller amounts of **I**, the pH should be adjusted to 2·7—2·8 by careful titration with 0·5N-HCl with the use of methyl orange as indicator; two drops of the acid are added per 30 ml. of solution, just after a red colour has been established. Under these conditions, neither the HCl nor the indicator can affect the **I** in the $Et_2O$-phase. P. S. ARUP.

**Correlation of different aspects of auxin action.** R. S. de Ropp and E. Markley (*Plant Physiol.*, 1955, **30**, 210—214).—Stem elongation took place only when the auxin (in agar) was applied to the apical end of the hypocotyl and was a max. with concn. of 0·01 to 0·1 $\mu$g. per ml. of agar. Max. increase in fresh wt. took place with 1 to 10 $\mu$g./ml. applied at the basal rather than at the apical end of the fragment. Initiation of roots was greatest with 0·1 $\mu$g./ml. applied apically. E. G. BRICKELL.

**Nature of the enzymically catalysed oxidation products of indolylacetic acid.** D. T. Manning and A. W. Galston (*Plant Physiol.*, 1955, **30**, 225—231).—The two most prominent oxidation products contain aromatic amino- and phenolic hydroxyl-groups and may resemble *o*-formamidoacetophenone in structure; hydroxy-*o*-aminoacetophenone is suggested as a possible product formed by hydroxylation of the benzene ring followed by oxidation of the pyrrole nucleus. E. G. BRICKELL.

**Plant growth-regulating substances. IX. Effects of pollen extracts and synthetic growth substances on the hop plant (*Humulus lupulus*, L.).** R. C. Seeley and R. L. Wain (*Ann. appl. Biol.*, 1955, **43**, 355—365).—Application of some synthetic plant growth-regulating substances markedly stimulated flowering laterals of the hop plant. The effect was specific to certain compounds and varied between varieties of hops and in the stage of flower development. The initial stimulation was not maintained by repeated application of the chemical. Untreated control laterals yielded a greater wt. of cones with a higher soft resin content. Application to flowering laterals of hop pollen extract and a flavonal glycoside obtained from the pollen failed to stimulate early cone development whether or not the laterals were protected from pollination. Striking changes in vegetative growth occurred when 2 : 3 : 5-tri-iodobenzoic acid was applied to mature or seedling hop plants, although the chemical had no effect on flowering comparable with that induced in tomato plants. A. H. CORNFIELD.

**Effect of growth-substances on growth and yield of wheat.** R. D. Asana, V. S. Mani and —. Vedprakash (*Physiol. Plant.*, 1955, **8**, 279—287).—Experiments on the effects of indol-2-ylacetic acid and $\alpha$-naphthylacetic acids applied to young wheat plants through the cut leaf-tips or by spraying indicate stimulation of growth, although the effects on the no. of ears and grains and on grain-size varied with the weather conditions during the five years over which the experiments extended. P. S. ARUP.

**Balance between free and bound growth-substance in germinating maize.** T. Hemberg (*Physiol. Plant.*, 1955, **8**, 418—432).—Distinction is made between free growth-substance [indolylacetic acid (**I**)] and bound growth-substance (**I**-precursor), the former being extractable from the ground grain by $Et_2O$ at 2° (with three changes of $Et_2O$) and the latter by $Et_2O$ at 28° during 45 hr. (with two changes). Contents of the precursor in maize decrease during storage for 3·5 months, whilst contents of **I** remain constant. Maize caryopses show increased contents of **I**, and decreased contents of the precursor after soaking in water, the effects being directly dependent on the time of soaking. Indications are obtained that the precursor occurs most abundantly in the endosperm, and that the conversion enzymes are located mainly in the embryo. Incubation of the maize during 3 hr. in aq. synthetic **I** increases the content of **I** in the kernel, but after 24 hr. most of the **I** has been converted into bound growth-substance. (28 references.) P. S. ARUP.

**Biochemical studies of micro-elements in green plants. I Deficiency symptoms in barley and changes in indolylacetic acid oxidase.** A. Fujiwara and M. Tsutsumi (*Tohoku J. agric. Res.*, 1954, **5**, 47—52).—Data obtained by growing barley seedlings in nutrients deficient in Fe, Mn, Zn, Cu, Mo or B are presented. Indolylacetic oxidase activity in the roots of the plants was not affected

appreciably by absence of Fe, Mn, Zn or B, but was markedly lowered by deficiency of Cu and (especially) Mo. Addition of Cu to the Cu-deficient nutrient restored the oxidase activity but corresponding replacement of Mo in the Mo-deficient medium was ineffective. Indolylacetic acid oxidase is probably a Cu-protein in the synthesis of which Mo plays a part. A. G. POLLARD.

**Physiology of plant roots. I. Influence of environmental conditions on growth of isolated roots of rice and wheat.** A. Fujiwara and K. Ojima (*Tohoku J. agric. Res.*, 1954, **5**, 53—61).—In tissue cultures of excised root tips, thiamine and pyridoxine accelerate growth in rice: in wheat pyridoxine was the more effective. Nicotinic acid had little action in this respect. A. G. POLLARD.

**Factors controlling meristematic activity in excised roots. VI. Effects of various anti-auxins on growth and survival of excised roots of *Lycopersicum esculentum*, Mill.** H. E. Street (*Physiol. Plant.*, 1955, **8**, 48—62; cf. *ibid.*, 1954, **7**, 212).—The effects of 2 : 3 : 5-triiodobenzoic acid (TIBA), α-(*p*-chlorophenoxy)-*iso*butyric acid, phenoxy*cyclo*propane carboxylic acid and 2-naphthoxyacetic acid (2-NOA) on excised tomato roots were unlike those of α-(1-naphthylmethylsulphide)-propionic acid (NMSP). The action of TIBA and 2-NOA resembled that of β-indolylacetic acid rather than that of NMSP. A. G. POLLARD.

**Apparent necessity of indolylacetic acid for growth of *Diploidia* (Fungi imperfecti).** A. C. Gentile and R. M. Klein (*Physiol. Plant.*, 1955, **8**, 291—299).—Observations on the growth of the fungus in a nutrient solution containing indol-2-ylacetic acid (**I**) and 2 : 4 : 6-trichlorophenoxyacetic acid (**II**) in various concn., separately or in admixture, demonstrate the toxicity of both substances and the counteracting of the toxicity of **II** by additions of **I**. The fungus synthesises **I** (in presence or absence of tryptophan); the toxicity of experimental additions of **I** can be explained by the assumption that such additions are in excess of the natural requirements of the fungus. Additions of **II** do not inhibit growth by affecting the biosynthesis of **I**, but (probably) by interfering with its action. The implications of these findings are discussed. (30 references.) P. S. ARUP.

**Effect of an anti-growth-substance on indolylacetic acid content in *Avena* coleoptiles.** P. Fransson and T. Ingestad (*Physiol. Plant.*, 1955, **8**, 336—342).—In the *Avena* straight growth test (cf. *ibid.*, 1953, **6**, 796) treatment (at the base) of the coleoptiles with $10^{-5}$ M-*p*-chlorophenoxy*iso*butyric acid reduces growth and increases the contents of indolylacetic acid in the coleoptiles, beyond the contents found in the untreated coleoptiles. Possibilities as to mechanism of the action of the anti-growth substance are considered.
P. S. ARUP.

**Aliphatic esters of indol-3-ylacetic acid. Preparation and activity in parthenocarpic fruit induction.** L. E. Weller, S. H. Wittwer and H. M. Sell (*J. Amer. chem. Soc.*, 1955, **77**, 4937—4938).—The following esters of indol-3-ylacetic acid are prepared from the acid and the appropriate alcohol, with an HCl catalyst: n-*hexyl*, m.p. 31—32°, n-*heptyl*, m.p. 26·5—27·5°, n-*octyl*, m.p. 27—28°, n-*nonyl*, m.p. 29·5—30·5°, n-*decyl*, m.p. 38·5—39·5°, n-*undecyl*, m.p. 39·5°, n-*dodecyl*, m.p. 48—49°, n-*tetradecyl*, m.p. 78—79°, n-*hexadecyl*, m.p. 60—62°, and n-*octadecyl ester*, m.p. 52°. All esters are biologically active in inducing parthenocarpic fruit development in the tomato, at a concn. of 1%. The activities, compared with that of the free acid, are: propyl to decyl esters, ~10 times as great; undecyl and dodecyl esters, equivalent; higher esters, less active.
O. M. WHITTON.

**Evaluation of the growth activity of naphthalene derivatives.** H. Burström (*Physiol. Plant.*, 1955, **8**, 174—188).—The comparative activities of various concn. of α- and β-naphthylacetic (1- and 2-NAA) and 1- and 2-naphthoxyacetic (1- and 2-NOAA) acids on wheat root growth are determined. 1-NAA and 2-NOAA act as auxins and 2-NAA and 1-NOAA as antagonists. Quant. effects of these acids probably depend only on the type of linkage between nucleus and side-chain whereas the qual. effects depend only on the position of the side-chain. A. G. POLLARD.

**Synthesis of 3-[3′-(5′-alkoxy(aryloxy)indolyl)]butyric acids.** N. N. Suvorov, V. P. Mamayev and L. B. Shagalov (*Dokl. Akad. Nauk SSSR*, 1955, **101**, 103—106).—The following 3-(3′-indolyl)-butyric acids have been prepared by a Fischer synthesis from the appropriate phenylhydrazones of Et 4-formylvalerate: 3-(5′-*methoxy*-, $C_{13}H_{16}O_3N$ (92% yield), m.p. 135—135·5°, 3-(5′-*benzyloxy*-, $C_{19}H_{19}O_3N$, m.p. 161·5—162·5°, 3-(5′-*phenoxy*-, $C_{18}H_{17}O_3N$ (29%), m.p. 107—108°, and 3-(7′-*chloro*-5′-*methoxy*-**indol-3′-yl)butyric acid**, $C_{13}H_{14}O_3NCl$ (31%), m.p. 135·5—136·5°. The root-stimulating activity of the first of these products was comparable to that of indolylacetic acid, whilst the remainder were relatively inactive.
R. TRUSCOE.

**Response of lettuce to spraying with maleic hydrazide.** R. S. Choudri and V. B. Bhatnagar (*Phyton*, 1955, **5**, 19—30).—Growth of cabbage lettuce sprayed, three weeks after transplanting, with aq. maleic hydrazide (0·05 to 0·20%) was inhibited. Three weeks after spraying, however, axillary buds grew out into flowering shoots (with 0·005% spray). The sprays prevented heading and with the onc. sprays premature death occurred. Sprays given to semi-mature heads tended to prevent "bolting". L. G. G. WARNE.

**Auxin–vernalisation relationships. I. Effects of certain synthetic auxins and their antagonists on vernalisation of *Brassica campestris*.** S. C. Chakravarti and V. N. K. Pillai (*Phyton*, 1955, **5**, 1—17).—Soaking of seed in aq. indolylacetic acid, indolylbutyric acid and naphthylacetic acid before vernalisation accelerated flowering more than did vernalisation alone. 2 : 4-Dichlorophenoxyacetic acid and tri-iodobenzoic acid reduced the effect of the subsequent vernalisation. L. G. G. WARNE.

**Effect of several chemicals on sprouting of stored table-stock potatoes.** H. Findlen (*Amer. Potato J.*, 1955, **32**, 159—167).—Treatment of Triumph and Pontiac potatoes with Me α-naphthylacetate (**I**) (0·017 g. per kg. of potatoes) gave very good control of sprouting during one month storage at 15°. The treatment was more effective when applied in aq. wax emulsion than in water. α-Chloronaphthalene (0·1 g. per kg. of potatoes) applied as an aq. wax emulsion prevented sprouting during storage but injured the tubers. α-Terpineol (0·05—0·20 g. per kg.) was much less effective than was **I** in preventing sprouting, whilst maleic hydrazide (0·015—0·090 g. per kg.) was ineffective. A. H. CORNFIELD.

**A form of *Actinomyces griseus* antagonistic to fungi.** M. Orsenigo, L. B. Orsenigo and R. Zucca (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 106—119).—A new strain of *Actinomyces griseus*, named UC53/1, shows marked antagonism to various fungi, but more towards bacteria. The active principle, which is insol. in common solvents, is not identical with grisein, actidione, streptomycin, or antibiotic 3510, which are produced by *A. griseus*.
F. R. PAULSEN.

**Production and identification of polyploids in red clover, white clover and lucerne.** A. E. Evans (*New Phytologist*, 1955, **54**, 149—162).—Treatment of seed with 0·2% aq. colchicine results in the production of polyploids in all these three species. Stomatal measurements serve to identify polyploid tillers and pollen grain measurements, polyploid inflorescences. Further, leaflet hairs are shorter in diploid than in tetraploid red clover plants.
L. G. G. WARNE.

**Application of the flame photometer and arc spectrograph in agronomic analyses.** M. Pinta (*Ann. Agron.*, 1955, **6**, 189—202).—The determination, in plant and soil extracts, of K, Ca, Na and Mg by flame photometry and of trace elements by arc spectrography are described and discussed. A. H. CORNFIELD.

**Analyses of plant materials using EDTA [ethylenediaminetetra-acetic acid] salts as solubilisers.** H. M. Bauserman and R. F. Olson (*J. agric. Food Chem.*, 1955, **3**, 942—946).—An extract, with a solution of an EDTA salt, of ground or pulped plant material contains the Na, K and Ca and some other metals, sugars, amino-acids and water-sol. org. compounds, and is suitable for analytical work. Sucrose may be determined using a saccharimeter, other carbohydrates and amino-acids by chromatographic separation and metals using flame spectrophotometric or gravimetric methods. (24 references.) E. M. J.

**Rapid determination of zinc in sugar cane leaf material.** H. Evans (*Trop. Agriculture, Trin.*, 1955, **32**, 142—146).—Leaf samples are taken from the middle portion of the lamina, avoiding the mid-rib, from the leaf showing the highest visible dewlap when the cane is 4½—5 months old. The leaf material is digested with $H_2SO_4$–$H_2O_2$. The digest is neutralised (KOH) to bromocresol green, and acetate buffer (pH 4—5) and aq. $Na_2S_2O_3$ (preventing interference from other metals) are added followed by dithizone in $CCl_4$. The $CCl_4$ layer is separated and the colour is matched in an absorptiometer (535 mμ). Results obtained are in reasonable agreement with those of the *Aspergillus* method. A. G. POLLARD.

**Determination of zinc in leaves and wood.** A. Bussmann (*Landw. Jahrb. Schweiz*, 1955, **69**, 365—381).—Selected methods, described in detail, include the following: two colorimetric methods based on the use of dithizone (accurate within ±3%), and a polarographic method (accurate within ±5%). In order to exclude interference by other metals, the use (in specified concn.) of Na thiosulphate and KCN is recommended in the colorimetric methods, and the use of KSCN in the polarographic method. (41 references.)
P. S. ARUP.

**Micro-determination of citric acid.** I. Reifer (*Acta biochim. polon.*, 1954/5, **1**, 293—305).—The method is based on the pentabromoacetone (**I**) reaction in presence of $CHCl_3$ (to facilitate separation of $KMnO_4$ and $H_2O_2$. Treatment of **I** with alkaline resorcinol

produces a red coloration. The method determines 0·5—40·0 $\mu g$ of citric acid with an accuracy of $\pm 5\%$ and is rapid and simple in operation. A. G. POLLARD.

**Determination of catalase activity in plant material.** B. Grabianowska (*Acta biochim. polon.*, 1954/5, **1**, 265—276).—Loss of catalase activity from samples of plant material during storage or during homogenisation prior to examination is temporarily prevented by addition of glucose + $CaCO_3$ + phosphate buffer (pH 7·0). Under these conditions the catalase activity may be retained for four days at 4° or for 24 hr. at 14°. A. G. POLLARD.

**Tentative method for sugar determination in laminæ; application to *Hevea brasiliensis*.** E. W. Bolle-Jones (*Physiol. Plant.*, 1955, **8**, 1—7).—Laminæ of the first or second whorl of sand-cultured *Hevea* seedlings were extracted with 80% ethanol and sugars in the extract were determined by a one-dimensional chromatographic method (described). The dominant sugar in the leaves was sucrose; smaller proportions of inositol and (in roughly equal proportions) glucose and fructose were also present. Deficiency of Mg in the nutrient increased the inositol content of the top whorl laminæ: B deficiency diminished the inositol content of the second whorl. The amount of sucrose in the first whorl was increased by lack of N and decreased by that of S, Ca, Fe or Mn. Deficiency of Ca also diminished the sucrose content of the second whorl. Glucose in the top whorl increased with lack of K whereas P deficiency increased the glucose and fructose contents in the second whorl. A. G. POLLARD.

**Separation of sugars and their derivatives by paper electrophoresis.** B. Galos and W. Ostrowski (*Acta biochim. polon.*, 1954/5, **1**, 171—184).—The separation is effected by use of borate complexes. Sugars which have closely similar $R_F$ chromatographically show considerable differences in rate of electrophoretic movement. The mobility of uronic acids was 1·5 × that of ketohexoses: that of hexoses approx. double that of corresponding methyl-hexoses and that of hexitols approx. half that of the corresponding sugars. A. G. POLLARD.

**Recovery and utilisation of tree extractives.** A. B. Anderson (*Econ. Bot.*, 1955, **9**, 108—140).—A review dealing with pine resins and turpentines, essential oils, tannins and dyes and a variety of tree exudates. L. G. G. WARNE.

**Crops and plant breeding.** H. W. Howard (*J. roy. agric. Soc.*, 1954, **115**, 126—137).—Recent developments in the breeding of new varieties of cereals, grasses, fodder crops and of disease-resistant potatoes are described. A. G. POLLARD.

## Crops and Cropping

**Effect on yield and leaf area of winter wheat of applying nitrogen as a top dressing in April or in sprays at ear emergence.** G. N. Thorne and D. J. Watson (*J. agric. Sci.*, 1955, **46**, 449—456).—Nitrogen, at the rate of 0·5 cwt. of N per acre, was given in April as Nitrochalk or as eight spray treatments with 2% aq. $NH_4NO_3$ applied to soil or to leaves in May–June. Similar increases in yield and N content of grain were obtained in all cases. The later treatments (May and June) caused smaller increases in leaf area index (leaf area per unit soil area) than did April dressings. The no. of shoots per in. of row and leaf area per shoot were increased by the April applications: later dressings affected only the leaf area per shoot. A single spray application of urea (20 gal. of 58% aq. urea per acre) in early June had a similar effect on yield as did the May–June sprays of $NH_4NO_3$. A. G. POLLARD.

**Effects of nitrogen applied at different dates and of other cultural treatments on eye-spot, lodging and yield of winter wheat.** G. A. Salt (*J. agric. Sci.*, 1955, **46**, 407—416).—With a long-straw (Squareheads Master 13/4) and a short-straw (Bersée) wheat the incidence of eye-spot and the extent of lodging diminished and yield increased when the rate of sowing was lowered from 3·0 to 1·5 bushels per acre. Spraying with $H_2SO_4$ (12·5% BOV, 100 gal./acre) produced qualitatively similar effects. Dressings of $(NH_4)_2SO_4$ decreased eye-spot infection if applied in Oct. but had little effect if given in Mar.–May. Straw yields and the amount of lodging were increased more by application of N in Mar.–April than by that in May. Effects of N on grain yields were similar for all dates of application when lodging was small, each increment of N producing successive increases in yield. Where the crop was almost completely lodged, Mar. and April applications of N diminished and those in May or Oct. increased yields, the amounts of fertiliser applied having little influence. A. G. POLLARD.

**Influence of fertilisers and manures on the content of phytin and other forms of phosphorus in wheat and their relation to soil phosphorus.** B. N. Srivastava, T. D. Biswas and N. B. Das (*J. Indian Soc. Soil Sci.*, 1955, **3**, 33—40).—Application of inorg. P increased the total P in wheat grain whilst N, K, org. manures and green manuring had no effect. Application of P, either alone or in combination with other fertilisers, increased phytin-, inorg.- and acid-sol.-P in the grain, whilst N and K had the reverse effect. Both total and available soil P were significantly correlated with all forms of P in the grain. Phytin-P in the soil was also significantly correlated with phytin-P in the grain. A. H. CORNFIELD.

**Inheritance and heritability of protein, niacin and riboflavin in oats.** K. J. Frey, H. H. Hall and M. C. Shekleton (*J. agric. Food Chem.*, 1955, **3**, 946—948; cf. J.S.F.A. Abstr., 1954, 121).—Data are presented on the inheritance and heritability of protein, niacin and riboflavin in three oat crosses. The genes determining high protein % could be either dominant or recessive, depending on the genetic background on which they operated. In two of the crosses the high protein % appears to be dominant; in the other cross low protein % seems to be dominant. High niacin and riboflavin contents appeared to be dominant in each of the crosses. Heritability % for protein and niacin contents ranged from 86 to 93; for riboflavin from 0 to 32. E. M. J.

**Relation between leaf-number and ear-development in spring-sown barley and oats.** S. Andersen (*Physiol. Plant.*, 1955, **8**, 404—417).—Observations by the author's method (cf. *ibid.*, 1952, **5**, 199) show small increases in ear-development in oats with increased rates of sowing, and larger corresponding increases in barley; the reverse tendency is observed for leaf-development. A decided negative correlation is found between the development of tillers and ear-development in the main shoot. Errors inherent in the leaf-counting method are examined; for the purpose of growth analysis, the method is satisfactory where great accuracy is not required; for greater accuracy, observations must be based on ear-development. P. S. ARUP.

**Influence of environment on seed and seedling mortality. I. Influence of time of planting on germination of maize. II. Pathogenic potential of the soil.** J. L. Harper, P. A. Landragin and J. W. Ludwig (*New Phytologist*, 1955, **54**, 107—118, 119—131).—I. Emergence of seedlings varies greatly with variety and sowing date. The differences are related to the time taken for emergence.

II. Maize grains placed in moist soil at low temp. (10 days at 5° or 8 days at 8°) and then transferred to a high temp. show heavy mortality. Mortality is greatly prevented by soil sterilisation or by coating the grains with a fungicidal dust. There is great variation in the results of different sampling dates. L. G. G. WARNE.

**Cereal foliage analysis. III. The nitrogen–yield relationship applied to maize. IV. Relationship between the nitrogen content of maize leaf and ear, yield, and plant density.** J. Dulac (*C. R. Acad. Agric. Fr.*, 1955, **41**, 500—504, 504—507).—III. Field experiments using maize indicate that there is a relationship between the N-content of ear and leaf and the yield of grain. The relationship holds in spite of considerable variation in yield, type of plant, density of plants and type of fertiliser applied. It is of use in indicating the best fertiliser and optimum plant density to be adopted.

IV. The density of planting and the N content of the soil are of great importance in determining yield (cf. Pt. III). N. M. WALLER.

**Nitrogenous manuring of rice: ammoniacal nitrogen and nitric nitrogen.** Th. Zaliki (*Feuill. agric., Soc. Publ. égypt.*, 1955, No. 86, Repr., 7 pp.).—Egyptian rice-field experience, contrary to current practice, has led to the conclusion that, whether the fertiliser is $NH_4$ sulphate or Chile nitrate, there is an optimum time of application, at about the 65th day after sowing in the case of direct culture, and about 90 days after sowing the nursery plants in the case of culture by transplantation, and that, provided the fertiliser is applied at this time, there is no difference between the yields obtained with the two fertilisers. J. S. C.

**Influence of night temperature and nitrogen nutrition on the growth, sucrose accumulation and leaf minerals of sugar beet plants.** A. Ulrich (*Plant Physiol.*, 1955, **30**, 250—257).—Ripening or "sugaring up" was induced by low night temp. and by N deficiency, the latter also decreasing top growth of the plants. E. G. BRICKELL.

**Effect of winter weather on the composition of fodder-beet left in the ground.** M. E. Castle (*J. Sci. Food Agric.*, 1955, **6**, 579—581).—When fodder-beet of high dry-matter content is left in the soil over the winter period, there is little or no change in chemical composition, palatability, or soundness. J. S. C.

**Potato planting trials.** M. Birecki and K. Kropkiewicz (*Roczn. Nauk rol.*, 1955, **70, A**, 515—547).—Effects of time of planting on the yield of tubers and of starch in different districts are examined. The relationship between starch yield (per acre) and sowing time is dependent on soil and climatic conditions during the period of max assimilation by the plants. A. G. POLLARD.

**Mixed planting of potato varieties.** K. Malec (*Roczn. Nauk rol.*, 1955, **70, A,** 549—555).—Considerable increases in yields of late potatoes resulted from planting alternate rows of late and early varieties. Yields of early varieties were not appreciably affected. A. G. POLLARD.

**Influence of period and depth of sowing on the growth, quality and yield of potatoes under various methods of cultivation.** M. Birecki, S. Behnke and J. Szulc (*Roczn. Nauk rol.*, 1955, **70, A,** 351—404).—Cultivation practices, e.g., sowing in ridges and on the flat, depth of planting, deep and shallow inter-cultivation are compared in light, heavy and peaty soil types. Production of starch in the tubers was favoured by ridging in light soils and by flat-planting in heavy soils and, in general, by fairly deep planting (10—15 cm.). Highest yields of tubers and of starch per acre were obtained by early sowing followed by ridging three times after the emergence of the plants. A. G. POLLARD.

**Potato planting rates under dryland culture in Western Nebraska.** H. O. Werner (*Amer. Potato J.*, 1955, **32,** 197—206).—Yields of three varieties of potatoes, grown without irrigation, from three planting rates (6, 12 or 24 bushels per acre) with two seed-piece sizes at two planting distances were studied over 3—7 years. The 6-bushel planting rate produced the greatest yields per bushel of seed planted, but yields per acre were less than with the higher planting rates. The 24-bushel rate produced somewhat greater yields than did the 12-bushel rate, but the increased returns from the higher rate were relatively small. The heavy rate was economical only when seed was cheap. With the intermediate planting rate close planting of small seed pieces gave better results than did wider planting of larger seed pieces. A. H. CORNFIELD.

**Effect of intensity and width of inter-row tillage on the yield of the potato crop.** M. A. Moursi (*Amer. Potato J.*, 1955, **32,** 211—214).—Yields of one variety of potato were slightly reduced, whilst those of another were unaffected, by inter-row tillage as compared with no tillage. Yields were not significantly different as between uncultivated, hand-weeded plots and cultivated plots, or between plots cultivated twice and those cultivated three times. Narrow inter-row tillage (8 in. from the plant) produced slightly higher yields than did wide inter-row tillage (6 in. from the plant), but only with one of the two varieties studied. The difference in response of the two varieties was ascribed to differences in the extent of lateral spread of the root system. A. H. CORNFIELD.

**Effect of different combinations of soil moisture and nitrogen levels on early plant development and tuber set of the potato.** G. A. Bradley and A. J. Pratt (*Amer. Potato J.*, 1955, **32,** 254—258).—Better top growth, tuber wt. and tuber set were obtained with high than with low moisture levels during early growth of potatoes. The level of applied N (0—180 lb. per acre) generally had little effect upon the no. of tubers set early in the season. A. H. CORNFIELD.

**Storage of ware potatoes in permanent buildings. II. Temperature of unventilated stacks of potatoes.** W. G. Burton, G. Mann and H. G. Wager (*J. agric. Sci.*, 1955, **46,** 150—163).—Immediately after harvest potatoes stored at 10° respired at a high rate, but this rate fell off to a constant value after about four weeks. Respiration increased when sprouting commenced. Over the storage temp. range (0—25°), the min. respiration rate occurred at about 5—7·5°. Data on heat production and dissipation in unventilated stacks of varying dimensions are presented and discussed. In general it is safe to store unventilated potatoes to a height of 6 ft. if mature and to 3 ft. if immature. If not stored very late, and if relatively clean, mature potatoes may be stored to 12 ft. Dirty potatoes should not be stored late. A. H. CORNFIELD.

**Mechanical damage to potatoes during harvesting and handling operations in the Red River Valley of Minnesota and North Dakota.** R. E. Nylund, P. Hemphill, J. M. Lutz and H. Sorenson (*Amer. Potato J.*, 1955, **32,** 237—247).—Field harvesting operations caused mechanical injury (cuts, bruises and cracks) to 26·4% of the potatoes handled, 10·1% being caused during digging, 0·5% during picking into baskets, 4·9% during the filling of field sacks, 5·7% during loading and hauling to the warehouse, and 5·2% during bin filling. During all these operations 15·6% of the skin was removed from the potatoes. Mechanical injury occurring during the grading of both washed and unwashed potatoes is also reported. A. H. CORNFIELD.

**Flavour tests on potatoes grown in soil where lindane (benzene hexachloride) was applied to cucumbers.** M. E. Kirkpatrick, G. S. Linton, B. M. Mountjoy and L. C. Albright (*Amer. Potato J.*, 1955, **32,** 259—264).—The effects of the previous year's application of lindane ($C_6H_6Cl_6$) at ~1·25 lb. per acre to the soil or foliage of cucumber plants on flavour of the tubers of five varieties of potato grown in the following year are reported. Bliss Triumph, Irish Cobbler and Pontiac had off-flavours, whilst Cherokee and Sebago had no off-flavours. Where off-flavours occurred these were somewhat worse in immature than in mature tubers. Storage for three months at 12·8° did not alter the extent of off-flavours. A. H. CORNFIELD.

**Grassland management. I. Cultivations, fertility, manuring and establishment.** H. G. Chippindale. **II. Management of grazing swards.** T. E. Williams. **III. Mixed farms: cattle and sheep.** W. E. Jones. **IV. General dairy farms.** O. G. Williams. **V. Dairy farms: arable dairying, including bail.** D. J. Columbus Jones. **VI. Feeding farms.** T. W. Evans. **VII. Hill sheep and cattle grazings.** A. J. Davies. **VIII. Grassland and crop potential.** W. Davies (*Minist. Agric. Fish.*, 1955, *Bull. No.* 154, 3—15, 15—24, 24—33, 33—40, 40—46, 47—51, 52—61, 62—66).—I. and II. are reviews. III. Rotation, choice of crops, choice of ley, seasonal growth and stock requirements are discussed, together with their applications to the farm. IV. Fundamental principles are reviewed and a general guide to grazing management tabulated. V. Permanent grass and leys are discussed with respect to their introduction into traditional arable districts. VI. The feeding of (i) spring-bought stores on summer grass, (ii) autumn-bought stores on summer grass, and (iii) home-reared stores on summer grass are reviewed. VII. Rotational fields and meadowland, enclosed lands and intakes, and mountain and hill grazings are discussed from the point of improving the productivity of nutritious food in the winter. VIII. A review. (22 references.) E. G. BRICKELL.

**Lucerne and lucerne–grass leys. I. Summer and autumn management of a lucerne–grass mixture grown on heavy land.** M. G. Barker, F. Hanley and W. J. Ridgman (*J. agric. Sci.*, 1955, **46,** 362—376).—Lucerne–cocksfoot leys were cut or grazed three times annually. Total yields of dry matter were similar whether cut or grazed although some differences in individual crops were apparent. Effects of grazing varied somewhat according to the time of year. Detailed effects of early and late defoliation are discussed. A. G. POLLARD.

**Lucerne and lucerne–grass leys. II. Nitrogenous manuring of a lucerne–cocksfoot ley.** W. J. Ridgman, F. Hanley and M. G. Barker (*J. agric. Sci.*, 1955, **46,** 441—448).—Effects of dressings of $(NH_4)_2SO_4$ (1 cwt. per acre, 1, 2 or 3 applications in Feb., June or Aug.) on the ley were compared on a heavy gault soil. Increased yields of grass were obtained without adverse effects on the growth of lucerne. Best results were obtained with the Feb. application. Recovery of fertiliser-N in the crop was poor. A. G. POLLARD.

**Effect of rate and frequency of phosphate application on pasture production.** F. W. Schaller and G. G. Pohlman (*West Virginia agric. Exp. Sta.*, 1955, *Bull.* 380, 17 pp.).—Increased yields of forage from permanent pasture were directly related to the amounts of superphosphate applied. The greatest increases in yield usually occurred in the second and third year after application. Liming increased forage yields on an acid soil and also increased the proportion of desirable species in the sward. There was little difference in response as between spring and autumn applications of superphosphate or lime. A. H. CORNFIELD.

**Intensive production of herbage for crop drying. VI. Effect of intensive nitrogen fertiliser treatment on species and strains of grass, grown alone and with white clover.** W. Holmes and D. S. MacLusky (*J. agric. Sci.*, 1955, **46,** 267—286).—A five-year manurial trial with various grasses and grass–clover mixtures, in which the herbage was cut 4—6 times annually, is recorded. The response of various grass species to N fertilisers and to culture in admixture with clover varied considerably, cocksfoot in pure culture and ryegrass and timothy in presence of clover giving the best return for N applied. Earliness, distribution of yield over the season, total protein yield and differences in performance between strains are discussed in detail. A. G. POLLARD.

**Digestibility and nutrient value of grass from the same source treated in different types of drying plant.** W. Schoch (*Landw. Jahrb. Schweiz.*, 1955, **69,** 343—356).—Tabulated details are given for the experimental conditions in eleven different drying processes, and for the nutrient values (as determined by feeding experiments on wethers) of the several products. In comparison with the nutrient value of the fresh grass, significant decreases in protein digestibility occurred in five of the driers. The important factors are the drying temp. and heat distribution in relation to the time of passage of the grass through the drier, and especially a suitable decrease in temp. towards the end of the drying. With short drying times (> 5 min.) moist grass can be treated without detriment with initial drying temp. of 600—700°. The moisture content of the dried product should be 10—12%. P. S. ARUP.

**Rhodes grass.** Anon. (*Qd agric. J.*, 1954, **78,** 71—80).—Climatic requirements, most suitable soils for growth, and methods of planting and management of Rhodes grass (*Chloris gayana*, Kunth.) are described. Because of its running habit the grass is particularly

suitable for soil conservation. The chemical composition (with respect to feeding value) of Rhodes grass is compared with that of buffel, Queensland blue, and Guinea grasses. Rhodes grass is high in protein when young, but protein content falls off rapidly with maturity. A. H. CORNFIELD.

**Recent advances in the conservation of forage crops.** M. J. Nash and S. J. Watson (*J. roy. agric. Soc.*, 1954, **115**, 20—26).—A short review devoted, mainly, to developments in silage making. A. G. POLLARD.

**Ensiling of lucerne with various preservatives.** J. H. Weniger and K. Funk (*Arch. Tierernähr.*, 1955, **5**, 33—40).—The effects of Kofa salt (Ca formate with a small % of $NaNO_2$), formic acid, and KCNS on the quality of ensiled young lucerne are comparatively evaluated by tests for pH, org. acids and digestibility. The effects of the preservatives are, in the above order, satisfactory, mediocre and unsatisfactory. (20 references.) P. S. ARUP.

**Magnesium deficiency in apple in British Columbia.** C. G. Woodbridge (*Canad. J. agric. Sci.*, 1955, **35**, 350—357).—Leaf blotch showed when the Mg content of the leaves was <0·18%; above 0·26% no symptoms were observed. Soil applications of Mg salts were ineffective but 2% aq. $MgSO_4$ as a foliage spray was beneficial. Jubilee and Newtown varieties showed most susceptibility to leaf blotch. E. G. BRICKELL.

**Chemical thinning of apples.** F. T. Bowman (*Agric. Gaz. N.S.W.*, 1955, **66**, 365).—Recommended strengths of naphthylacetic acid for thinning different varieties of apple are given. A. H. CORNFIELD.

**Water relationships of the fruit and leaves as related to "hard-end" of the Bartlett pear.** W. B. Ackley (*Wash. agric. Exp. Sta.*, 1954, *Tech. Bull.* 15, 35 pp.).—The water contents and water deficits of leaves and the osmotic concn. of leaves and fruits of Bartlett pears growing on (*a*) Japanese rootstock, *Pyrus serotina*, on which "hard-end" pears have frequently been found and (*b*) French stock, *P. communis*, on which affected pears are rarely found, were compared. The leaves of the trees on the French rootstock had a higher water content and lower water deficit than did those of trees on the Japanese rootstock. Although leaves and fruit from trees on Japanese rootstock had a higher osmotic concn. than had comparable samples from French rootstock trees, osmotic gradients between fruit and leaves were similar for trees on both rootstocks. Water imbalance in the tree may be the cause of "hard-end." A. H. CORNFIELD.

**Winter hardiness of stone fruit varieties.** H. W. Foyle and F. L. Overley (*Wash. agric. Exp. Sta.*, 1954, *Bull.* 553, 19 pp.).—There were varietal differences in the degree of winter hardiness of peach, apricot, plum and cherry trees. Hardy and non-hardy varieties of each species are listed. A. H. CORNFIELD.

**Thinning grapes with naphthylacetic acid.** E. El-Din Farrag (*Trop. Agriculture, Trin.*, 1955, **32**, 147—149).—Grapes were effectively thinned by spraying, at full bloom, with 0·05% naphthylacetic acid. The average wt. per berry was increased but the average wt. per bunch was substantially unchanged. A. G. POLLARD.

**Foliar nutrition of vines.** E. A. Asriev (*Vinodelie i Vinogradarstvo*, 1954, No. 3, 45—48).—Repeated spray applications of N, P, K, B and Mn in various combinations to leaves of 18-year-old vines increased the sugar content of the grapes in nearly all cases, P and B giving the best results. The acidity of the juice was not greatly affected. The wt. per bunch, wt. per berry and juiciness were improved especially by P + K. Max. effects on yield were produced by N + P + K + Mn + B whereas N + P + K did not affect yields. Addition of appropriate mineral salts to Bordeaux mixture applications is recommended. HORT. ABSTR. (A. G. P.).

**Artificial colouring and ripening of fruits.** C. D. Stevenson (*Qd agric. J.*, 1954, **78**, 151—155).—The artificial colouring and ripening of bananas, citrus, papaws and tomatoes by the $C_2H_4$, $C_2H_2$, coal gas and paraffin burner methods are described. A. H. CORNFIELD.

**Selection of carrots for carotene content. II. Sub-normal content at low temperatures.** O. Banga, J. W. de Bruyn and L. Smeets (*Euphytica*, 1955, **4**, 183—188).—Carrots grown at low temp. have a narrow long conical root and a low carotene content. L. G. G. WARNE.

**Occurrence of molybdenum deficiency in cauliflower in West Germany, and remedial measures.** E. Brandenburg and C. Buhl (*Z. PflKranhh.*, 1955, **62**, 514—528).—The deficiency (of which the symptoms are described) occurs in many localities, especially on soils of moderate or high acidity, and is aggravated by applications of peat or fertilisers of acid reaction. The effects can sometimes be mitigated by liming, but satisfactory results can generally be obtained only by applications of Na molybdate (mixed with sand or in solution) at 1—4 kg. per hectare or 2—3 g. per cu. m. of soil. The health of affected plants can be restored by such applications. The increased incidence of the disease is probably (in part) due to increased cultivation of the deficiency-sensitive "Alpha" varieties of cauliflower. Such varieties are useful indicator-plants for the deficiency. (25 references.) P. S. ARUP.

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**Early fruiting and boll maturity of cotton as affected by sodium and root aëration.** F. L. Selman and R. D. Rouse (*Soil Sci.*, 1955, **80**, 281—286).—Cotton plants were grown in sand irrigated with complete nutrient solutions but with variable amounts of Na and K. The roots were either kept well aërated by allowing the nutrient solution to drain away, or poorly aërated by keeping the roots immersed until the next irrigation. During the early blooming stage bolls were set only on the plants receiving Na and with poorly aërated roots. After 40 days plants on all other treatments began to set bolls. In a second test the nutrients contained additional P and Cl. After 35 days the plants receiving Na were taller, and the poorly aërated plants had longer lateral branches than had the well-aërated. After five months this aëration effect was reversed. Bolls on the poorly aërated plants matured, and vegetative growth ceased 30 days before that in aërated plants. At 110 days the no. of blooms and bolls was greater when Na was included and the roots were poorly aërated than when the plants were well aërated. The K level did not affect this. Na increased the percentage of blooms set without regard to aëration at high but not at low or adequate levels of K. T. G. MORRIS.

**The biting taste of Burley tobacco. I. Relations between the biting taste and chemical components.** A. Fujiwara and M. Kurosawa (*Tohoku J. agric. Res.*, 1954, **5**, 229—237).—Leaves grown in the colder climate of the north island of Japan, and having an undesirable biting flavour, were characterised by high total and protein-N and low carbohydrate contents and by a narrow ratio of reducing sugar to total N. Leaves with a wider ratio were of satisfactory flavour. A. G. POLLARD.

**Mineral nutrition of sugar cane in British Guiana. I. Survey of nutrient status of cane using rapid fresh-tissue tests.** H. Evans (*Trop. Agriculture, Trin.*, 1955, **32**, 124—133).—Data for the contents, in the cane, of K, P, Ca, Mg, Mn, $SO_4''$ and Cl (by Morgan's reagent), N (by visual symptoms), Fe, Zn, Cu and Mo (by HCl extraction), Al (by staining with hæmatoxylin) are presented and discussed. A. G. POLLARD.

**Rapid analysis of sugar-cane stalks for determinations of cane quality.** H. Evans and F. Le Grand (*Trop. Agriculture, Trin.*, 1955, **32**, 134—141).—Sample canes are cut longitudinally from top to bottom by a circular saw so arranged that the sawdust is directed to a chute and collected. The dust is macerated with water in a Waring-type blender (3 min.), dry Pb acetate (Horne) is added and the blender is operated for a further 3 min. The mixture is filtered and sugar in the filtrate is determined polarimetrically. A. G. POLLARD.

**Some aspects of the nutrition of tea in southern India.** V. Jayaraman and P. de Jong (*Trop. Agriculture, Trin.*, 1955, **32**, 45—47).—In a 12-year experiment the crop yields were increased by application of N fertiliser [40—80 lb. of $(NH_4)_2SO_4$ per acre] and, over the later years of the trial, by smaller dressings of KCl (20—40 lb. per acre). Deficiency of K in the soil increased the mortality of bush owing to their inability to survive pruning operations. A. G. POLLARD.

**Copper: its occurrence and rôle in tea leaf.** R. Child (*Trop. Agriculture, Trin.*, 1955, **32**, 100—106).—Although no specific Cu-deficiency disease was apparent in the growing bush the polyphenol oxidase involved in the fermentation of the leaf is a Cu-protein. Unsatisfactory fermentation is associated with leaves containing Cu <12 p.p.m. The mechanism of the adverse effect of Cu deficiency is discussed. A. G. POLLARD.

**Cacao propagation in Malaya with special reference to cacao nursery technique.** C. Whitehead (*Malayan agric. J.*, 1954, **37**, 203—210).—Methods used are described. A. H. CORNFIELD.

**Hagotan for the extraction of Manila hemp fibre.** D. S. Boyce (*Malayan agric. J.*, 1954, **37**, 218—224).—A simple mechanical contrivance (Hagotan) for the extraction of Mamila hemp fibre, where only moderate quantities of hemp are available, is described. A. H. CORNFIELD.

**Biological and technicological properties of Early White Przebędowo lupin.** S. Barbacki, S. Jankowski and K. Latawiec (*Roczn. Nauk rol.*, 1955, **70, A**, 479—513).—This lupin variety, originally bred from a bitter stock, is a long-day type, responding readily to vernalisation. Early sowing favoured wt. per 1000 seeds, high fat and low alkaloid contents (0·04—0·08%). Milling yielded 35% of flour suitable for use in baking and about 50% of groats. When mixed (3—5%) with wheat and rye flour, lupin flour affected pastry in a manner similar to that of soya-bean. In bread the loaf vol. was

lowered. The amino-acid distribution of the lupin protein is compared with that of other leguminous seeds. A. G. POLLARD.

**Effect of farmyard manure and superphosphate on berseem yield and nodulation and on the nitrogen and available phosphorus contents of the soil.** S. Sen and S. S. Bains (*J. Indian Soc. Soil Sci.*, 1955, **3**, 41—48).—Application of farmyard manure or superphosphate to berseem increased yields of both green fodder and seed in proportion to the amounts of P applied (16—24 lb. $P_2O_5$ per acre). When applied on the basis of equivalent amounts of P, superphosphate was more effective than farmyard manure. High levels of applied P were necessary to maintain yields of berseem over three years. Nodulation on berseem roots and N content, particularly of the 0·6-in. layer, of the soil increased with amount of P applied. Available soil P was increased to a greater extent when P was applied as superphosphate and farmyard manure than when applied as superphosphate alone. A. H. CORNFIELD.

**Replanting oil palms.** J. D. Ferwerda (*Trop. Agriculture, Trin.*, 1955, **32**, 45—47).—Effects of felling the old stand of palms before planting anew and also of fertiliser treatment are examined. Best development of the new stand was obtained by completely felling the old stand and applying a suitable complete fertiliser. When the old stand was retained the mortality of young trees was high. A. G. POLLARD.

**Changes in soil properties associated with the growth of cactus (*Opuntia dilleni*).** A. Sen and D. Singh (*J. Indian Soc. Soil Sci.*, 1955, **3**, 23—27).—In comparison with adjacent non-vegetated soil, soils growing cactus had lower clay, base-exchange capacity and pH, higher total N content and salinity, and unchanged org. matter content. A. H. CORNFIELD.

**Safflower—production, processing and utilisation.** P. F. Knowles (*Econ. Bot.*, 1955, **9**, 273—299).—A semi-popular account with an extensive bibliography. (129 references.) L. G. G. WARNE.

**Loofah—the sponge gourd.** W. M. Porterfield, jun. (*Econ. Bot.*, 1955, **9**, 211—223).—The botany, cultivation and uses of this plant are dealt with. Analyses of the fruit and of the seed oil are given. L. G. G. WARNE.

**Regeneration of *Chlorophora excelsa*, (Mvule), in Uganda in relation to soil-root conditions.** G. H. S. Wood and E. M. Chenery (*E. Afr. agric. J.*, 1955, **21**, 34—41).—The surface soil up to 50 ft. from the boles of large Mvule trees invariably had a high pH (6·2—7·0) and base status. Reasons for this are discussed. The root system of the tree is extensive and has a particularly wide radius when ironstone pans restrict downward growth. Under natural conditions poor regeneration is due mainly to poor soil physical conditions. A. H. CORNFIELD.

**Development of seed production and the seed trade in Europe (Project No. 214).** (European Productivity Agency, O.E.E.C., Paris, 1955, 138 pp.). **Seed production in Sweden.** E. Akeburg (17—24). **Present-day status of seed production, testing and distribution in the O.E.E.C. participating countries.** G. Nilsson-Leissner (25—36). **The preliminary study of new varieties of cultivated plants from the point of view of the authorities.** R. Mayer (37—46).. **Testing of new varieties from the point of view of the plant breeder.** A. Akerman and O. Tedin (47—59). **The need for uniform variety trials in participating countries.** A. Sandison (61—70). **The standardisation of official lists of varieties in the participating countries.** R. Milatz (71—82). **The desirability of a uniform international terminology in relation to seed certification schemes.** A. Kjaer (83—90). **Aspects of import and export regulations.** N. H. H. Addens (91—95). **Plant quarantine regulations in international seed trade.** E. Gram (97—101). **Possibilities of seed production and multiplication in the most suitable areas of the participating countries.** M. Thielebein (103—110). **Plant breeders' rights.** G. Weibull (111—123).—Papers read at the international conference on seed production in Stockholm, July 12th—17th, 1954. J. S. C.

**Durability of softwood cuttings in Polythene film.** G. Krüssmann (*Dtsch. Baumsch.*, 1954, **6**, 271—272).—Cuttings of pear, *Symphoricarpus, Syringa, Ribes alpinum* and *Lonicera pileata*, defoliated and packed as for export, but with an extra wrapping of Polythene, and stored for 35 days remained fresh and showed good callusing and some root formation. HORT. ABSTR. (A. G. P.).

## Pest Control

**Method of studying active mycelia on living roots and other surfaces in the soil.** J. L. Harley and J. S. Ward (*Trans. Brit. mycol. Soc.*, 1955, **38**, 104—118). L. G. G. WARNE.

**Disinfestation of soil by heat, flooding and fumigation.** A. G. Newhall (*Bot. Rev.*, 1955, **21**, 189—250).—A review. L. G. G. WARNE.

**Fungi that attack microscopical animals.** C. L. Duddington (*Bot. Rev.*, 1955, **21**, 377—439).—A review containing a section dealing with the use of these fungi to control eelworms in the soil. A bibliography of 131 references. L. G. G. WARNE.

**Control of plant diseases by use of antagonistic organisms.** R. K. S. Wood and M. Tvert (*Bot. Rev.*, 1955, **21**, 441—492).—A review with an extensive bibliography (190 references). L. G. G. WARNE.

**Some responses of fungi to light.** W. D. McLellan, H. A. Borthwick, I. Bjornsson and B. H. Marshall, jun. (*Phytopathology*, 1955, **45**, 465).—Responses to light by eight out of the 13 fungi under observation include wide variations in one or more of the following functions: sporulation, sclerotial, sporodochial or pycnidial production, zonation or pigmentation. P. S. ARUP.

**Cause of difference in sensitivity to DDT as between the third and fourth instar larvæ of the potato beetle (*Leptinotarsa decemlineata*, Say).** R. Langenbuch (*Z. PflKrankh.*, 1955, **62**, 564—572).—The fourth instar larva of the beetle is much less sensitive to dry DDT than is the third instar, which in comparison with the fourth, has a considerably lower lipin content (on live wt. and per ml. of hæmolymph) and fewer hæmocytes. The absorbed DDT is, therefore, nearer to saturation point in the body-lipins of the third than in those of the fourth instar. These observations accord with the previous findings (cf. *ibid.*, 1953, **60**, 168) that for solutions of equal concn. of a lipin-sol. insecticide, the toxicity to insects is inversely proportional to the solvent power of the solvent employed. (21 references.) P. S. ARUP.

**Insecticidal activity and chemical constitution: analogues and isosteres of DDT.** A. Stringer, D. Woodcock and E. J. Skerrett (*Ann. appl. Biol.*, 1955, **43**, 366—378).—A no. of isosteres of DDT and related compounds were examined for insecticidal activity using the grain-weevil, locust and cotton stainer. Replacement of the Cl atoms of DDT by Me and OH groups resulted in marked loss of activity. In the nitroalkane series the optimum structure for max. toxicity was present in 1 : 1-di-*p*-chlorophenyl-2-nitropropane. Results are discussed in the light of current theories of DDT structure and activity. A. H. CORNFIELD.

**Comparison between effective mechanism of phenyl mercury borate [Merfen] and that of mercuric chloride on lower fungi.** H. Frank (*Zbl. Bakt.*, 1955, II, **108**, 660—671).—On the spores of five representative fungi on a suitable medium, Merfen is lethal at far lower concn. than equiv. amounts of $HgCl_2$, PhOH, or mixtures of these. Merfen inhibits (mainly) the respiration and glucose-uptake of yeast, whilst $HgCl_2$ inhibits N-uptake. Merfen is absorbed by yeast cells in smaller amounts than is the case with $HgCl_2$ and has a wider range of enzymic inhibitory action, which includes enzymes containing SH-groups. Merfen is absorbed by yeast-cells in smaller amounts than is $HgCl_2$; its lethal effect is a direct function of its "toxic partial pressure"; any such connexion is much less marked in the case of $HgCl_2$. (26 references.) P. S. ARUP.

**The *Daphnia* test for detecting contact insecticides.** W. Pfaff (*Z. PflKrankh.*, 1955, **62**, 361—370).—The use of *Daphnia* to detect minute amounts of insecticide residues and, in some cases, by observations of specific movements to distinguish between different types of insecticides, e.g., chlorinated hydrocarbons and phosphoric esters, is described. Data obtained with 10 different insecticides are given. A. G. POLLARD.

**Standardisation of air-flow in insect suction traps.** L. R. Taylor and W. S. Coleman (*Ann. appl. Biol.*, 1955, **43**, 390—408).—The air-flow and its variations in 16 insect suction traps of five different types are reported. A. H. CORNFIELD.

**Combined apparatus for sterile filtration and delivery of filtrate in measured amounts.** H. Stolp (*Zbl. Bakt.*, 1955, II, **108**, 656—659).—The advantages of sterilisation of nutrient media by filtration are reviewed. The apparatus (on the German market) described, consists of a vessel in which the liquid passes by suction through a membrane-filter, a 1-l.-graduated reservoir from which larger deliveries of the filtrate can be made, two burettes (100 and 50 ml.) for smaller deliveries, and a vac. pump. The apparatus, including all connexions, is in glass; all parts coming into contact with the liquid can be heat sterilised before use; the delivery-jet is shielded by a bell-shaped cover. The apparatus has functioned satisfactorily during continued use. P. S. ARUP.

**[Measurement of] the crystallisation rate of Bordeaux mixture.** H. P. Burchfield and J. Schechtman (*Contr. Boyce Thompson Inst.*, 1955, **18**, 215—223).—Bordeaux mixture deteriorates, within a few hours, the amorphous hydrogel changing to the crystalloid state.

The degree of deterioration of a 10—10—100 Bordeaux mixture (**I**) was examined by addition of sucrose solution, which rapidly dissolved freshly-prepared material but only slowly dissolved aged material owing to the decreased sp. surface of the crystalloid. It was found that deterioration proceeded in three stages, viz., (*a*) an initial induction period of low deterioration, which probably corresponds with the time required for nuclei to form, (*b*) a period of rapid deterioration, and (*c*) a period of slow deterioration, attributed to increase in size, and corresponding decrease in surface available for reaction of the particles formed. The induction period for **I** at 45° is 23 min. (10 references.) J. S. C.

**Infra-red determination of Dieldrin and DDT in mixtures.** G. E. Pollard, W. M. Saltman and P. Yin (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 478—482).—The Dieldrin and DDT are extracted in a chromatographic column containing Celite by means of $CS_2$. The absorbence of the solution in $CS_2$ is determined at 10·96 and 14·06 $\mu$., and the amounts of the two substances present are calculated by means of an equation derived from measurements on the two substances separately. A. A. ELDRIDGE.

**Colorimetric determination of ethyl 4 : 4-dichlorobenzilate (Chlorobenzilate) as a spray residue.** H. J. Harris (*J. agric. Food Chem.*, 1955, **3**, 939—941).—A modification of the Schechter–Haller procedure is described for the determination of residual Chlorobenzilate on such crops as apples, pears, etc. The compound is nitrated and made to react with Na methylate; a red complex is formed which is measured spectrophotometrically at 538 m$\mu$. Any DDT present in the spray is dechlorinated with alcoholic KOH and extracted with light petroleum. The Chlorobenzilate is saponified to the K salt of 4 : 4′-dichlorobenzilic acid, and the acid obtained. After nitration the same coloured compound is formed with Na methylate as given by the ester. There was a gradual drop in concentration of residues on the apples during a two-month period after application of the spray. E. M. J.

**Evaluation of fungicidal action of basic chlorides of copper and calcium.** R. Radoni and S. di Caro (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 198—202).—The action of mixed oxychlorides of Cu and Ca was tested against conidia of *Alternaria* by the method of R. Ciferri and E. Baldacci (*Atti Ist. bot. Univ. Pavia*, 1943, **I**, Ser. 5, 106). The biological efficiency per unit wt. of Cu increased with Ca content until the latter reached ~7·2%, beyond which the biological action fell very sharply. F. R. PAULSEN.

**Evaluation of *OO*-dialkyl S-alkylthiomethyl phosphorodithioate.** Elton L. Clark, G. A. Johnson and E. L. Mattson (*J. agric. Food Chem.*, 1955, **3**, 834—836).—The chemical and biological properties of a new series of phosphorodithioates as systemic insecticides are described. An appropriate *OO*-dialkyl hydrogen phosphorodithioate is treated with formalin, and a mercaptan. The resulting product tested against the spotted spider mite (*Tetranychus bimaculatus*) was most effective if the phosphorodithioate radicle is ethyl, compared with methyl, *iso*propyl, or *n*-propyl, and the mercaptan radicle is ethyl or *iso*propyl. (13 references.) E. M. J.

**Determination of salts of ethylenebisdithiocarbamic acid in presence of copper salts in fungicides.** P. Fontana and R. Martelli (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 187—191).—Org. matter is decomposed in presence of ferrocyanide, which inactivates the Cu. A sample, containing 0·2—0·3 g. of the ethylenebisdithiocarbamate, together with ~1 g. of $K_4Fe(CN)_6$, is digested with 50 ml. of boiling 2N-$H_2SO_4$ for ~30 min. The first absorption vessel contains 25 ml. of 15% Pb acetate, and the second 25 ml. of methanolic KOH. Further details follow the method originally suggested by D. G. Clarke *et al.* (*Analyt. Chem.*, 1951, **23**, 1842). F. R. PAULSEN.

**Three important pests of cereals in Britain.** H. W. Miles and M. Miles (*J. roy. agric. Soc.*, 1954, **115**, 47—59).—The distribution of eelworm, wheat bulb fly and wheat shoot fly, the mode of attack on the crop and control measures are discussed. A. G. POLLARD.

**Soil-borne diseases of cereals.** M. D. Glynne (*J. roy. agric. Soc.*, 1954, **115**, 41—46).—Factors influencing the incidence and severity of the diseases are reviewed. A. G. POLLARD.

**Effect of the " water-soak " seed treatment on the germination of certain barley varieties grown at different locations.** D. C. Arny and C. Leben (*Phytopathology*, 1955, **45**, 518—519).—The " water-soak " treatment of barley seed for the control of loose smut caused appreciable reduction in germinative capacity, the effect varying considerably among varieties and between different locations. A. G. POLLARD.

**Seasonal incidence of *Sporobolomyces* on cereal leaves.** F. T. Last (*Trans. Brit. mycol. Soc.*, 1955, **38**, 221—239).—Five colonies were found on cereal leaves until they had lived half their lives, and the colonies reached a max. after the leaves died. More were found on dead leaves in summer than in winter. In the presence of *Tilletiopsis* the number of *Sporobolomyces* colonies was reduced, especially on barley. L. G. G. WARNE.

**Control of the blue oat mite, *Penthaleus major* (Duges).** A. W. S. May (*Qd agric. J.*, 1954, **78**, 201—202).—Characteristics of the mite, which attacks cereals and groundnuts, are described. The pest is controlled with DDT (0·25 lb. per acre suitably diluted in the form of a spray or dust). A. H. CORNFIELD.

**Effects of calcium sulphamate and sodium sulphanilate on small grains and on stem rust development.** A. A. C. Livingston and J. E. Livingston (*Phytopathology*, 1955, **45**, 503—506).—Ca sulphamate (9—12 lb. in 5 gal. of water per acre), applied to cereal crops during pollination and repeated after seven days, lowered the subsequent germination of wheat and oat seed but not that of barley. The incidence of stem rust was diminished but the effects on grain yields were not consistent. Na sulphanilate also diminished stem rust and increased wheat yields without injury to the grain. Best results were obtained by applications made one week after tillering and again six days after flowering. A. G. POLLARD.

**Toxin production by *Helminthosporium victoriae*.** H. H. Luke and H. E. Wheeler (*Phytopathology*, 1955, **45**, 453—458).—Filtrates from cultures of the fungus on the medium described show at dilutions >1 p.p.m. pathogenicity (as measured by retardation of root elongation) towards non-resistant varieties of oats, but not towards resistant oats, other cereals, or garden vegetables. The symptoms produced by the filtrates are characteristic of those produced *in vivo*. Five mutant cultures produced from a single-spore isolate are described; three of these are pathogenic in varying degrees, whilst two are non-pathogenic. The culture filtrates can be stored or autoclaved without loss of pathogenicity at pH <4, but not at higher pH. P. S. ARUP.

**Effect of potash fertilisers on certain fungal diseases of rice.** M. Orsenigo (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 8—21).—Rice with K deficiency was stunted, highly sterile, and had short panicles and light grains. " Stem rot " due to *Sclerotium oryzæ* produced high sterility, but was much reduced by adequacy of K. " White-tip," caused by *Aphalenchoides oryzæ*, did not increase sterility, and was increased by high K. " Brown-spot " (*Helminthosporium oryzæ*) did not increase sterility and was unaffected by K levels. Infection by " white-tip " and " brown-spot " simultaneously resulted in high sterility and reduced grain wt. at all K levels, while " brown-spot " with " stem-rot " reduced plant height. F. R. PAULSEN.

**Control of snow-fungus (*Fusarium nivale*, Ces.) of winter rye on high ground by fungicidal dusts and sprays applied in late autumn.** F. Wagner (*Z. PflKrankh.*, 1955, **62**, 539—544).—Mercurial seed-dressings are ineffective against the fungus under prolonged snow-cover but applications of " Brassicol Super " dusting power at 50 kg. per hectare gave satisfactory protection under snow-cover lasting for 83 days. In greenhouse experiments with infected seed, pentachloronitrobenzene gave very satisfactory, and $C_6H_6Cl_6$ fairly satisfactory control. P. S. ARUP.

**Seed disinfection. XI. Control of head smut [*Sorosporium reilianum*, (Kuhn) McAlpine] of maize.** H. Jacks and G. J. Graham (*N.Z. J. Sci. Tech.*, 1955, **37**, 141—145).—Dusting maize seed in the field with material based on org. Hg, Dichlone and thiram, significantly reduced the head smut. R. H. HURST.

**Changes in rice caryopses due to seed-borne fungi.** M. Orsenigo (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 22—33).—In work on fungus-inoculated rice seeds, *Curvalaria* was non-pathogenic, but *Helminthosporium oryzæ* and *Fusarium* spp. produced infection in seedlings. With the last-named, activity decreased with rise in temp., but at 28° typical symptoms of " bakanae disease " appeared. Both *Curvalaria* and *Fusarium* caused discoloration of kernels, but there was no infection of the hulls. (29 references.) F. R. PAULSEN.

**Control of *Verticillium* wilt of potatoes in Connecticut, 1954.** P. E. Waggoner and G. S. Taylor (*Amer. Potato J.*, 1955, **32**, 168—172).—Application of a foliage spray of 2 : 4-D-Na salt (1000 p.p.m.) 40 days after planting decreased the severity of *Verticillium* wilt in two out of four tests. The plants were distorted by the treatment, but recovered and made rapid growth. Yields of tubers were usually reduced by the treatment. Severity of wilt was not affected by four weekly sprays of Terramycin HCl (500 p.p.m.), HD 469 (a promising chemotherapeutic agent against *Fusarium* wilt of tomatoes; 1000 p.p.m.), or seed or soil treatment with zineb. Treatment of seed potatoes with zineb, Semesan Bel, Manzate, Vancide 51, Lo-738 (polyethylenebis-thiuram trisulphide), dimethyldithiocarbamic acid-Na salt, or 2-mercaptobenzthiazole-Na salt had no effect on incidence of wilt or yield of tubers. A. H. CORNFIELD.

**Reactions of potato varieties to late blight and insect injury as reflected in yields and percentage solids.** F. J. Stevenson, R. V. Akeley and R. E. Webb (*Amer. Potato J.*, 1955, **32**, 215—221).—In a year unfavourable for development of late blight, yields of tubers and % of dry matter in the tubers of four resistant varieties compared favourably with those of two susceptible varieties. In a bad blight year unsprayed resistant varieties produced relatively high yields whilst unsprayed susceptible varieties produced very poor yields. When sprayed with basic Cu every seven days, yields from both resistant and susceptible varieties were increased, but yields from the latter were still much the lower. The % of dry matter in the tubers of all varieties was increased by the basic Cu sprays. Spraying with DDT improved yields of some of the varieties. A. H. CORNFIELD.

**Effect of some soil factors on efficiency of fungicides in controlling *Rhizoctonia solani*.** M. H. K. Rushdi and W. F. Jeffers (*Phytopathology*, 1955, **45**, 466).—Complete inhibition of the fungus in a liquid medium is achieved by Puratised Agricultural Spray at 4 p.p.m., and by Arasan, Semesan, Vanicide 51 or Zerlate at 20 p.p.m. Fermate and Act-dione at 20 p.p.m. cause 75% inhibition. Dithane and Cu compounds are practically ineffective. In soil, increases in moisture or temp. do not affect fungicidal efficiency; increases in pH increase, decrease or fail to affect the efficiency of the several fungicides. Additions to the soil of 1% of starch, hay, or straw decrease the efficiency of nabam or Puratised Agricultural Spray; additions of 5% of hay reduce the efficiency of all the prep. except Vanicide 51. P. S. ARUP.

**Cytochemistry of leaves of potato infected with "yellow mosaic."** F. M. Gerola and M. Grilli (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 136—142).—Cytochemical studies on mesophyll of the leaves of the potato var. Majestic infected with "yellow mosaic" virus have demonstrated changes in nuclear dimensions, chromatin distribution, polymerisation of deoxyribonucleic acid, content of ribonucleic acid, and nucleolar vol. (28 references.) F. R. PAULSEN.

**Caryology of potatoes affected with virus diseases.** F. M. Gerola and M. Grilli (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 143—151).—Potato plants var. Majestic affected by various virus diseases show characteristic vol. decreases in nucleus and nucleolus, a high nucleus to nucleolus ratio, decreased ribonucleic acid in cytoplasm and plastids, and increased polymerisation of the deoxyribonucleic acid. All these are signs of reduced metabolism. F. R. PAULSEN.

**New strain of the potato leaf-roll virus.** R. E. Webb (*Amer. Potato J.*, 1955, **32**, 173—179).—The characteristics of the new strain are described. A. H. CORNFIELD.

**Indicator plant for a strain of potato virus Y.** K. Silberschmidt and E. Rostom (*Amer. Potato J.*, 1955, **32**, 222—227).—Potato virus Y (*ibid.*, 1954, **31**, 213—217) produced characteristic local lesions and systemic symptoms on the leaves of *Nicandra physaloides* (L.) Gaertn. A. H. CORNFIELD.

**Virus latent in potato plants.** R. H. Bagnall and R. H. E. Bradley (*Amer. Potato J.*, 1955, **32**, 252—253).—Evidence that potato sap often contains a latent virus other than those commonly known to infect potato plants is presented. A. H. CORNFIELD.

**Diagnostic host for potato virus A.** R. E. Webb and R. W. Buck, jun. (*Amer. Potato J.*, 1955, **32**, 248—252).—Characteristic local lesions, distinctly different from those incited by the other major potato viruses, appeared on leaves of *Solanum demissum* when inoculated with potato virus A. A. H. CORNFIELD.

**Cultivation of *Phytophthora infestans* (Mont.) de Bary on artificial media, and antibiotic action of various organisms against the fungus.** E. Magdon (*Zbl. Bakt.*, 1955, II, **108**, 703—716).—Improvements in the Feustel–Schönbrunn method of the cultivation of the fungus on potato-root press-juice malt-agar include inoculation of freshly-poured plates with single conidia from fresh cultures on potato-slices, and incubation at 22° and at R.H. 98%. A large no. of the (73) strains of *Actinomycetes* tested show, in streak cultures, widely varying inhibitory effects on the mycelial growth, but no effects on the spore-germination of the fungus. Similar effects are observed for a smaller proportion of the (30) soil bacteria tested. For the *Actinomycetes*, three different types of zonal inhibition are observed. A few of the bacteria promote the growth of the *Actinomycetes*, which, in their turn, can then inhibit the former. P. S. ARUP.

**Causes for differences between resistance of sugar-beet varieties to *Cercospora beticola*, Sacc.** A. Kovács (*Phytopath. Z.*, 1955, **24**, 283—298).—Spores of the fungus germinate more slowly on the leaves of resistant than on those of non-resistant varieties. Rain, dew or sprayed water collected from young or old beet-leaves contains germination- or growth-inhibiting material, but greater inhibitory effects are observed for the liquid collected from the leaves of resistant than that from the leaves of non-resistant varieties. Similar differences are observed for water extracts (obtained by immersion of the leaves at room temp.) of the different varieties. Analogous effects are observed for the germination of the spores of *Alternaria tenuis*. P. S. ARUP.

**Effect of fungicides on seedling diseases of legumes and grasses in Saskatchewan.** H. W. Mead (*Canad. J. agric. Sci.*, 1955, **35**, 329—336).—In sterile soil inoculated with *Fusarium culmorum*, treated seed of lucerne, red clover and crested wheat grass grew better than untreated seed but in general the response of legume and grass seed to treatment varied considerably because of difference in seed samples and soil conditions. Seed treatment does not control post-emergence blighting. E. G. BRICKELL.

**Comparison of forage yields from row-grown and sward pastures.** J. H. Teakle (*Qd agric. J.*, 1954, **79**, 17—23).—Over four years a green panic grass pasture grown in rows on an alluvial soil yielded 45% more hay than when grown as a sward. A pasture of alternate rows of panic grass and lucerne yielded an additional 25% of hay. The nutritive value of the row grass, even without lucerne, was superior to that of the sward grass. A. H. CORNFIELD.

**Toxicities of certain insecticides to the sweet-clover weevil, *Sitona cylindricollis*, Fahr. (Coleoptera: Curculionidæ), and the protection of seedling crops.** W. R. Allen and W. L. Askew (*Canad. J. agric. Sci.*, 1955, **35**, 344—349).—Toxicity of Dieldrin, heptachlor and parathion to sweet-clover weevil exceeded that of DDT, Malathion and toxaphene in laboratory tests. Under field conditions Dieldrin and heptachlor at 0·5 lb. per acre gave greater protection to sweet-clover seedlings than did parathion, DDT or Metacide, and increased both the no. of plants surviving and the dry wt. per plant at harvest. E. G. BRICKELL.

**Virus diseases and the fruit farmer.** R. V. Harris (*J. roy. agric. Soc.*, 1954, **115**, 83—97).—The occurrence, dissemination, effects and control of virus diseases of soft and tree fruits are discussed briefly. For effective control a continuous supply of virus-free stock is essential and the strict elimination of diseased stock must be practised throughout the industry. A. G. POLLARD.

**Presence in air of ascospores of *Venturia inæqualis*, (Cke.) Wint.** G. Govi (*Ric. sci.*, 1955, **25**, 2098—2102).—Three methods for collecting ascospores of *V. inæqualis* in the foliage of apple trees are discussed. Inspiration of air by a pump placed among the leaves caused maceration of the spores by dust simultaneously inspired. Direct counting from greased plates placed at various heights in the tree was interfered with by dust and vegetable fragments. By placing 5 sq. cm. portions of leaf in petri dishes over water the expelled spores could be collected centrifugally. Counts by the last-named method in relation to rainfall, ambient temp. and tree growth are considered. T. P. MCLAUGHLIN.

**Substitutes for lead arsenate and DDT in the apple spray programme.** W. S. Hough and C. H. Hill (*Virginia agric. Exp. Sta.*, 1954, *Bull.* 467, 18 pp.).—DDT was more effective than $PbHAsO_4$ in controlling codling moth, but did not always control the red-banded leaf roller or the plum curculio. In the laboratory parathion, Malathion, Metacide and EPN were more effective than was DDT in killing young codling moth larvæ within three days after application; methoxychlor was less effective than was DDT, but was more effective than $PbHAsO_4$. In cage tests parathion and Malathion were highly effective as contact sprays against codling moth and as residual inscticides for at least one week following application. Residual effects were greater than that of DDT. Methoxychlor was as effective as DDT as a contact spray but had poor residual effects. Dilan, EPN and heptachlor were fairly good contact poisons but showed poor residual effects. Parathion was the only material tested which exhibited egg-killing properties that may be of practical importance against codling moth eggs. A. H. CORNFIELD.

**Captan, zineb, and Captan + zineb for control of fruit-rot phase of *Botryosphæria ribis* on apples.** R. R. Romanko and J. W. Heuberger (*Phytopathology*, 1955, **45**, 466).—Sprayings with the above prep. gave 76—83% reductions in the incidence of the rot, Captan or Captan + zineb being more effective than zineb alone. The control effects persisted during storage of the apples at 1·5°. P. S. ARUP.

**Control of the pear and cherry slug, *Caliroa limacina*.** E. H. Zeck (*Agric. Gaz. N.S.W.*, 1955, **66**, 294—295).—The pear and cherry "slug," the larvæ of *Caliroa limacina*, which also feeds on plums, quinces and hawthorns, is controlled with 0·1% DDT or $PbHAsO_4$ (1 lb. per 80 gal.) sprays. A. H. CORNFIELD.

**Biology, life history and methods of control of the leaf-curling plum aphid, *Brachycaudus helichrysi*, (Kltb).** S. H. Bennett (*J. hort. Sci.*, 1955, **30**, 252—259).—A detailed study of the aphid, bred on damson, reveals that hatching and reproduction occur before bud-opening. Satisfactory control can be obtained by spraying the tree

during dormancy with DDT or $\gamma$-$C_6H_6Cl_6$ (the latter at 0·13%). The latter is also effective after leaf-curling has occurred due to attacks by the aphid. P. S. ARUP.

**Injury to deciduous fruit trees by an ectoparasitic nematode (*Xiphinema* sp.); a promising control measure.** R. E. Adams (*Phytopathology*, 1955, **45**, 477—479).—Application of BHC (960 lb. per acre) controlled the nematode which injures the roots of peach and apple and is the probable cause of "pout" in apples. The treatment improved the growth of the trees and did not impart abnormal flavours to the fruit. A. G. POLLARD.

**Use of zinc ethylenebisdithiocarbamate against *Peronospora*.** P. Fontana and G. Zampighi (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. i, 176—186).—The fungicidal film produced by Zn ethylenebisdithiocarbamate on vine seedlings was removed by heavy rains, but otherwise proved stable. The fungicide was detected in leaflets produced after the treatment, i.e. it was translocated in the plant. The efficacy of the Zn compound against *Peronospora* infections was equal to that of Cu salts, but decreased if mixtures with the latter were used. F. R. PAULSEN.

**Greenhouse testing of fungicides against *Botrytis* rot (grey mould) of strawberry and other soft fruits.** M. H. Moore and R. P. Tew (*J. hort. Sci.*, 1955, **30**, 213—219).—The test consists in dipping ripening soft fruit in solutions of the fungicides and, after air-drying, spraying with spore suspensions (containing a wetting agent) of the fungus; the fruits are then incubated in a moist chamber. Thiram, Captan and salicylanilide proved more efficient (at concn. of 0·5% of active ingredient) than were several other fungicides, thiram giving the best results. The use of 10% dusts proved less successful. P. S. ARUP.

**Control of *Botrytis* rot (grey mould) of strawberries, and effects of fungicide spray residues on processed fruit.** R. W. Marsh, J. T. Martin and A. Crang (*J. hort. Sci.*, 1955, **30**, 225—233).—Three sprayings with Captan (at 0·25% of active material) or thiram (at 0·4%) reduced the rot by ~67%; at 0·125%, Captan was less effective; Tecnazene or salicylanilide were ineffective. The max. residues of Captan found on ripe strawberries (amounting to 19 p.p.m.) did not affect the flavour or ascorbic acid content of the fresh fruit. Thiram residues gave marked, and Captan residues moderate tainting in canned strawberries. In the latter case, the effects were due to decomposition products of Captan. No tainting occurred in jams made from the sprayed fruits. P. S. ARUP.

**Strawberry *Botrytis* rot (grey mould) control: a field test of Captan at East Malling.** A. H. M. Kirby, M. H. Moore and Dorothy J. Wilson (*J. hort. Sci.*, 1955, **30**, 220—224).—In trials extending over two years, a suspension of Captan (2% of 50% wettable powder) was applied in two post-blossom sprayings at 700 gal. per acre, strawed alleys being sprayed by lance. The sprayings increased the marketable crop by 30%, and were most effective on irrigated plots, where losses in the absence of spraying were greater than on non-irrigated plots. P. S. ARUP.

**Effect of environmental factors on the appearance of "June Yellows" in strawberries and its significance for the development of a test method.** J. P. Braak (*Euphytica*, 1955, **4**, 189—196).—"Yellows" in Climax strawberries is not affected by photoperiod or fluctuations in temp around 5°. Exposure to low temp. accelerates subsequent development of "yellows" whilst at high temp. (20°) symptoms of "yellows" may disappear and the newly produced leaves are green. L. G. G. WARNE.

**Strawberry ripe fruit rot.** O. W. Sturgess (*Qd agric. J.*, 1954, **78**, 269—270).—Characteristics of the ripe fruit rot, due to a *Glœsporium*, are described. Routine fungicidal treatments used for leaf-spotting fungi will check the rot. Application of a Cu fungicide to the runner beds in the autumn is also helpful. A. H. CORNFIELD.

**Citrus bud mite *Aceria sheldoni*, (Ewing) Eriophyidæ, in Kenya.** R. le Pelley (*E. Afr. agric. J.*, 1955, **21**, 22—24).—The occurrence and distribution of the mite which was first recognised on grapefruit and lemon trees in 1955, but which is now known to be widespread, are described. Of several materials tested for control of the pest, 0·1% Chlorbenzilate has given the best results. A. H. CORNFIELD.

**Control of red scale, *Aonidiella aurantii*, and yellow scale, *A. citrina*.** P. C. Hely (*Agric. Gaz. N.S.W.*, 1955, **66**, 376—379).—Characteristics of the two scales are described. They are controlled by fumigation with HCN under cover or by spraying with white oil (1 gal. of 80—90% oil emulsion per 40 gal. water), 0·075—0·100% parathion, or 0·15—0·20% Malathion. A. H. CORNFIELD.

**Control of black scale *Saissetia oleæ*.** E. H. Zeck (*Agric. Gaz. N.S.W.*, 1955, **66**, 374—375).—The black scale is controlled by spraying with white oil emulsion (4 fl. oz. per gal.) by fumigation with HCN. A. H. CORNFIELD.

**Control of the Queensland fruit fly, *Dacus (Strumeta) tryoni*.** Anon. (*Agric. Gaz. N.S.W.*, 1955, **66**, 29—32).—The range of plants attacked by the Queensland fruit fly and methods of treatment or disposal of waste and infected fruit are described. Foliage baits are described. A. H. CORNFIELD.

**Control of greenhouse thrips, *Heliothrips hæmorrhoidalis*.** E. H. Zeck (*Agric. Gaz. N.S.W.*, 1955, **66**, 375, 381).—The thrips, which attack many ornamental shrubs and fruit trees, are controlled by spraying with 0·1% DDT or nicotine sulphate (1 fl. oz. per 4 gal.). A. H. CORNFIELD.

**General-purpose garden pesticide.** C. K. Dorsey and M. E. Gallegly (*West Virginia agric. Exp. Sta.*, 1954, *Tech. Bull.* 365, 25 pp.).—Nine different vegetable crops were sprayed and dusted with various mixtures of the fungicides zineb, Maneb and Captan and the insecticides methoxychlor, DDT and Malathion. Although a no. of combinations controlled all the pests encountered, it was concluded on the basis of yield data, cost and toxicity factors that the zineb–methoxychlor–Malathion combination (2 : 2 : 2·5 lb. per 100 gal. spray or 3·9% : 5·0% : 4·0% dust) was the most generally satisfactory. A. H. CORNFIELD.

**Foot rot of tomatoes induced by *Phytophthora capsici*.** P. D. Critopoulos (*Bull. Torrey bot. Club*, 1955, **82**, 168—182).—This fungus was isolated from tomatoes affected with foot rot in California and the addition of mycelium to the soil around the stems of healthy tomato plants induced the disease to develop. It was pathogenic also to bean, pepper and pea plants. L. G. G. WARNE.

**Effect of nutrient sprays on *Fusarium* wilt of tomato.** J. R. Bloom and J. C. Walker (*Phytopathology*, 1955, **45**, 433—444).—Spraying of tomato foliage with aq. urea before inoculation wuth *Fusarium oxysporum* f. *lycopersici*, (Sacc.) Snyd & Hans. retarded disease symptoms, whilst spraying after inoculation increased the symptoms; the effects were ∝ the concn. (1—4M) of the urea. Sprays containing KCl or NaCl produced no effect when applied before, but increased the symptoms when applied after inoculation. Aq. $CaCl_2$ was ineffective by itself, but when applied in conjunction with KCl it counteracted the above described effect of the latter. $NaH_2PO_4$, Na glycerophosphate and Mg salts were ineffective. P. S. ARUP.

**Efficacy of two soil fumigants and two insecticides against root-knot nematode in tomato and okra.** K. L. Olsen and N. F. Thomas (*Turrialba*, 1954, **4**, 23—28).—Fumigation of soil with Dow-fume W-40 or D-D (30 gal. per acre in both cases) controlled the nematode. Neither parathion or Aldrin was effective at the tested rates of application. HORT. ABSTR. (A. G. P.).

**Incidence and control of cauliflower mosaic in South-West England.** J. G. Jenkinson (*Ann. appl. Biol.*, 1955, **43**, 409—422).—Yields of broccoli were increased and extent of infection by cauliflower mosaic was much reduced when plants were raised in seedbeds at least 0·5 mile from old infected plants. Barrier crops (kale or barley) reduced the extent of infection somewhat. The manner of spread of the disease in the field is described. Plants infected as seedlings produced little or no curd, whilst those infected near maturity yielded almost as well as did uninfected plants. Movement of alate aphids was positively correlated with the no. of infected plants. Visual symptoms in field plants generally occurred 8—9 weeks after infection. A. H. CORNFIELD.

**Control of French bean fly.** P. C. Hely (*Agric. Gaz. N.S.W.*, 1955, **66**, 92—93).—The French bean fly, *Agromyza phaseoli*, was very effectively controlled with 0·05% DDT sprays applied weekly, after two initial sprays at 4-day intervals, up to blossoming. $\gamma$-$C_6H_6Cl_6$ (0·044% initial sprays followed by 0·022% sprays) also gave fairly good control. Although 0·01% E605 and HETP (1 : 1200) appeared to control the fly and gave good yields of beans, stem infestation was high. A. H. CORNFIELD.

**The onion fly, *Hylemyia antiqua*, in Denmark: biology and control measures.** J. Jørgensen (*Tidsskr. Planteavl.*, 1955, **59**, 252—279).—The life cycle of the fly is described. Seed dressing with $Hg_2Cl_2$ or DDT did not give satisfactory control. Very promising results were obtained with chlordane, used as a seed dressing, as a viscous solution in which plants were dipped before planting-out in the field or as a 10% prep. applied directly to the soil. When used as a seed dressing chlordane (up to 125 g. per kg. of seed) did not injure germination. $C_6H_6Cl_6$, at the rate of 20 g. per kg. of seed, delayed germination and suppressed the growth of the seedlings. A. G. POLLARD.

***Botrytis* leaf spot on onions and its control.** O. T. Page (*Canad. J. agric. Sci.*, 1955, **35**, 358—365).—Spotting and wilting of leaves by *B. squamosa*, Walker, is described. Manzate, Parzate (zineb), Vancide F-995W or Orthocide 50W as foliar sprays, give control. Downy mildew [*Peronospora destructor*, (Berk.) Casp.] is also destroyed. E. G. BRICKELL.

**Antibiosis in relation to pink root of shallots.** T. E. Freeman and E. C. Tims (*Phytopathology*, 1955, **45**, 440—442).—Of two soils in the same field, one (Mhoon silty clay) was heavily infested with *Pyrenochæta terrestris*, whilst the other (Sharkey clay) was not infested. The latter soil showed, in comparison with the former, higher actinomycetes counts, comprising a higher proportion of forms antagonistic to the fungus; these antagonistic forms reduced the virulence of the fungus towards onion seedlings. Infected seedlings were more prone to attack in sterilised than in non-sterilised soil. P. S. ARUP.

**A new hyaline species of *Verticillium*, *V. intertextum*, sp. nov.** I. Isaac and R. R. Davies (*Trans. Brit. mycol. Soc.*, 1955, **38**, 143—156).—Although isolated from a wilting Japanese maple this fungus (diagnosis given) did not induce wilting either in maples or in other plants tested. In culture max. growth is at 25° and pH 7·2. Sucrose, dextrose, maltose and glycerol are all utilised well, whilst $NaNO_3$ is a better source of N than $(NH_4)_2SO_4$, $NH_4NO_3$, asparagine or peptone. L. G. G. WARNE.

**Control of the emperor gum moth, *Antheræa eucalypti*.** E. H. Zeck (*Agric. Gaz. N.S.W.*, 1955, **66**, 34—35).—The caterpillar of the emperor gum moth, which feeds on gum, brush box and pepper trees is controlled with 0·1% DDT spray. A. H. CORNFIELD.

**Control of the pine weevil (*Æsiotes leucurus*) and the cypress pine buprestid beetles (*Diadoxus erythrurus* and *D. scalaris*).** E. H. Zeck (*Agric. Gaz. N.S.W.*, 1955, **66**, 295—296).—The females are deterred from laying eggs by painting the trees with bluestone paint ($CuSO_4.5H_2O$ 1·5 lb.–$Ca(OH)_2$ 1·5 lb.–water 2 gal.). A. H. CORNFIELD.

**Influence of water content of poplar on its resistance to *Cytospora chrysosperma* (Pers.) Fr.** H. Butin (*Phytopath. Z.*, 1955, **24**, 245—264).—Laboratory experiments with poplar twigs of uniform size and varying moisture content show a direct connexion between the extent to which the twigs have been dried, and the success of the infection by the fungus through surface wounds. The threshold range of loss of moisture as a % of the original moisture content is 18—22. Losses of moisture above this critical range are found by microscopical observation to interfere with the normal mechanical defence reactions which build up barricade tissue. Positive infection results can be counteracted by restoring the moisture deficit in the twigs. Channels for infection are afforded by the normally functioning vascular tissue of leaves, but not by vessels which have been blocked by lignification preceding leaf-fall. P. S. ARUP.

**Chemical composition of leaves of diseased tobacco: ring spot, bacterial wilt and *Fusarium* wilt.** F. A. Wolf and F. T. Wolf (*Phytopathology*, 1955, **45**, 506—508).—Infection, with ring spot increased the total and protein-N, and decreased the total sugar content of the leaves. Bacterial wilt had similar effects and also increased the light-petroleum extractives. *Fusarium* wilt resulted in diminution in sugars and an increase in the petroleum extract. A. G. POLLARD.

**Tobacco anthracnose. II. Physiological characteristics of the causal organism in relation to epidemiology.** E. A. Riley (*Trop. Agriculture, Trin.*, 1955, **32**, 150—155).—The organism, *Colletotrichum tabacum*, Böning, grew best at pH 7·0 and was favoured by high R.H. Certain soil organisms are probably antagonistic to *C. tabacum*. Recommended measures for controlling anthracnose include, weeding by hormone herbicides and spraying with Bordeaux mixture or synthetic org. fungicides, the timing of the sprays being of primary importance. A. G. POLLARD.

**Tobacco eelworm investigations in Uganda.** T. E. T. Trought (*E. Afr. agric. J.*, 1955, **21**, 28—31).—The root-knot eelworm is found throughout the tobacco-growing areas of Uganda as well as in other areas. Treatment of the seedbed with D-D reduced infection of seedlings from 75% to 22%. The degree of infestation of the plants at harvest was reduced when seedbeds and/or field sites were treated with D-D. Yields of green leaf were increased by about 75% when seedbeds were treated, but were not affected when field sites, but not the seedbeds, were treated. A. H. CORNFIELD.

**Nutrition and metabolism of the tobacco wilt, *Fusarium*.** F. T. Wolf (*Bull. Torrey bot. Club*, 1955, **82**, 343—352).—*Fusarium oxysporum* var. *nicotianae* utilises fructose, xylose and maltose very well, and ribose, mannose, glucose, galactose, sucrose, trehalose, cellobiose, raffinose, dextrin and melezitose satisfactorily. Arabinose, lactose and melibiose are poorly utilised. $NH_4^+$, $NO_3^-$ and amino-acid-N can all be used, but aspartic acid, glutamic acid and alanine are better as sources of N than $NH_4^+$ or $NO_3^-$. Of 34 nucleic acid derivatives none was as effective a N source as were amino-acids. Ethyl alcohol is produced when glucose is utilised. In cultures a pigment rubrofusarin is produced, but fusarinic acid could not be isolated from the cultures. L. G. G. WARNE.

**Laboratory observations on the effects of insecticides on the white coffee borer beetle, *Anthores leuconotus*, Pasc.** R. Foster (*E. Afr. agric. J.*, 1955, **21**, 6—9).—Dieldrin and Endrin were more effective than were DDT and chlordane in causing knock-down of the white coffee borer beetle when applied to the tarsi. Males and females responded in a similar fashion. The effect of a given dosage was similar whether applied to one tarsus or distributed between two tarsi. A. H. CORNFIELD.

**Bark diseases of coffee.** G. B. Wallace and M. M. Wallace (*E. Afr. agric. J.*, 1955, **21**, 25—27).—A general account of coffee bark and scaly bark diseases of coffee together with methods for preventing infection and for control. A. H. CORNFIELD.

**Methyl bromide gas treatments of dormant gladiolus corms and of sclerotia of *Sclerotium rolfsii*.** T. R. Carpenter and E. T. Gammon (*Phytopathology*, 1955, **45**, 520—521).—Corms and sclerotia were treated with various concn. of MeBr (3—20 lb. per 1000 cu. ft.) at 27° for 4 hr. With dosages of 3 lb. emergence from corms was unaffected; with 5—10 lb. treatments emergence was reduced to 0—10% but emerging plants showed stimulative effects; the 15—20-lb. treatments prevented all emergence. Sclerotia were unaffected by treatments up to 10 lb., showed a survival rate <50% with the 15-lb. dosage and were completely killed by the 20-lb. treatment. A. G. POLLARD.

**Fusarium disease of *Gladiolus*.** E. W. Buxton (*Trans. Brit. mycol. Soc.*, 1955, **38**, 193—201).—*Fusaria* isolated from rotting gladioli corms can cause "yellows" in the growing plants and isolates from yellowed plants can induce rots in stored corms. Both diseases are due to *F. oxysporum*, Fr. f. sp. *gladioli* (Massey) Snyder and Hansen. L. G. G. WARNE.

**Sclerotia production in *Stromatinia gladioli* as affected by potassium and other inorganic nutrients.** B. H. Marshall, jun. (*Phytopathology*, 1955, **45**, 465).—In cultures of the fungus on mineral–sucrose solutions, the no. of sclerotia formed is greatly increased by increases in K, appreciably increased by increases in Mg or P, but hardly affected by the concn. of B in the nutrient. The size, but not the no. of sclerotia is directly influenced by the concn. of N. P. S. ARUP.

**Host specialisation of *Erisiphe cichoracearum* from *Zinnia*, phlox and cucurbits.** J. A. Schmitt, jun. (*Mycologia*, 1955, 688—701).—*E. cichoracearum* isolated from *Zinnia elegans* would infect other species of *Zinnia* as well as species of many other genera. Isolates from some other Compositæ were rather more specialised. An isolate from *Phlox drummondii* was specific for this host, and an isolate from *Cucurbita pepo* infected only species of this family. L. G. G. WARNE.

**Browning and blackening of plant tissues. III. Occurrence in the leaves of *Dahlia* and several other plants of chlorogenic acid as principal browning agent.** M. Shiroya and S. Hattori (*Physiol. Plant.*, 1955, **8**, 358—369; cf. *ibid.*, 63).—The leaves of *Dahlia* and seven other plants were extracted with boiling water, and the extracts were subjected to a procedure involving pptn. with Pb acetate and extraction with ether by means of which three fractions were obtained as colourless solutions. The fractions all gave a brown coloration when oxidised by means of polyphenoloxidase. The presence of chlorogenic acid (**I**) as the principal browning agent was demonstrated in all the leaves, and that of caffeic acid in the leaves of *Dahlia*, *Clematis* and *Viburnum*. The enzymic oxidation of **I** was partly inhibited by ascorbic acid. In leaves subjected to atm. oxidation at 60° during 5 min. caffeic acid and **I** disappeared completely, glutamic acid disappeared almost completely, whilst alanine contents increased considerably. Citric acid and tyrosine were isolated from *Dahlia* leaves. P. S. ARUP.

**A *Helminthosporium* stem rot of cacti.** R, H. Durbin, L. H. Davis and K. F. Baker (*Phytopathology*, 1955, **45**, 509—512).—Symptoms of the disease are described. Control measures include sterilisation of the soil (by steam or chemicals) and application to the plants of Captan (2 lb. per 100 gal.) with a spreader-adhesive prep. A. G. POLLARD.

**Control of scurf of sweet potatoes.** T. J. Nugent, H. T. Cook and L. L. Harter (*Virginia Truck Exp. Sta.*, 1954, *Bull.* 113, 10 pp.).—The characteristics of scurf (caused by *Monilochætes infuscans*) of sweet potatoes are described. The relative effectiveness of selection of scurf-free seed potatoes, seed-potato treatment, sprout treatment and use of vine or sprout cuttings as control measures were compared. Selection of scurf-free seed was not practicable. Seed treatment with Hg compounds and/or S was usually ineffective. Treating sprouts with S dust or Semesan Bel reduced scurf significantly. Over three years scurf index was much lower in plants grown from sprout cuttings than in plants grown from pulled sprouts. A. H. CORNFIELD.

**Virus diseases of cacao in West Africa. IX. Strain variation and interference in virus 1A.** A. F. Posnette and J. McA. Todd (*Ann.*

*appl. Biol.*, 1955, **43**, 433—453).—Further investigations into strain variation and interaction in the virus 1A group are described. A. H. CORNFIELD.

**Prevalence of two species of *Cercospora* on groundnuts.** J. S. Hemmingway (*Trans. Brit. mycol. Soc.*, 1955, **38**, 243—246).—*C. personata* appeared late in the season on groundnuts in Tanganyika, but increased so rapidly that it became a more serious disease-producing factor than *C. arachidicola*. L. G. G. WARNE.

**Transmission of groundnut rosette virus.** H. H. Storey and A. K. Ryland (*Ann. appl. Biol.*, 1955, **43**, 423—432).—Transmission characteristics of the disease by *Aphis craccivora*, Koch, are described. A. H. CORNFIELD.

**Sudden death disease of cashew trees in Tanganyika.** G. B. Wallace and M. M. Wallace (*E. Afr. agric. J.*, 1955, **21**, 42—43).—A preliminary note recording the presence, in the Southern Province of Tanganyika, of a disease of cashew nut trees (*Anacardium occidentale*), which is very similar to and probably identical with the "sudden death" disease of the clove tree in Zanzibar. Some characteristics of the disease are described. A. H. CORNFIELD.

**Tannin- and phenol-decomposing enzymes of *Endothia parasitica*.** G. Bazzigher (*Phytopath. Z.*, 1955, **24**, 265—282).—Esterase activity by the fungus is demonstrated by means of two-dimensional paper chromatography with respect to tannin and gallic esters *in vitro*, and in infected tissue of *Castanea sativa*. Tannin, which is toxic to the fungus, is converted into less toxic products. The enzyme is formed by the fungus on a nutrient medium in the presence, but not in the absence of tannin. The presence in cultures of the fungus of a peroxidase and of a polyphenoloxidase of the laccase type is also demonstrated; enzymes of these types also occur in the wood of *C. sativa*; polyphenolase activity is stimulated as a response to wounding. The characteristics of the above enzymes are investigated, and their probable functional interrelationships are discussed. The course of the decomposition of the tannins of *C. sativa* by the enzymes of the fungus is shown by two-dimensional chromatograms. The breakdown is promoted by the wounding of the tissue, and is much more complete *in vivo* than *in vitro*. P. S. ARUP.

**Epidemiology and control of Nosema disease of the honey-bee.** L. Bailey (*Ann. appl. Biol.*, 1955, **43**, 379—389).—The epidemiology of the disease, due to *Nosema apis* (Zander), is described. Satisfactory control was obtained by sterilising old broodless combs by fumigation with HCHO or $CH_3COOH$, the latter being preferable since it does not poison honey or pollen in the combs. A. H. CORNFIELD.

**Comparative herbicidal action of the synthetic growth-substances 2 : 4-D and MCPA.** O. Schmidt (*Mitt. biol. Zentralanst. Berlin-Dahlem*, 1954, No. 77, 119 pp.).—Plants susceptible to both 2 : 4-D and MCPA reacted similarly to both, though often more rapidly to MCPA. Weeds usually propagated by seed were most susceptible in the earliest stages whereas those propagated by roots showed max. sensitivity just before flowering or at the time of most active longitudinal growth. A fall in temp. during or immediately after treatment prolonged or inhibited the response. High R.H. after application increased and rainfall lowered the herbicidal action. Soil, nutritional and light conditions also influenced the effects. The action of a given amount of herbicide per unit area was but little affected by the concn. of the spray in which it was applied. The susceptibility of numerous weed species to 2 : 4-D and MCPA is recorded. HORT. ABSTR. (A. G. P.).

**Herbicidal action of chloropicrin (Larvacide).** H. Hähne (*Z. PflKrankh.*, 1955, **62**, 612—617).—In treating soil with chloropicrin at the optimum temp. (16—22°) weed destruction near the surface is sometimes much less satisfactory than at lower levels. These effects (not observed for treatments at lower temp.) are attributable to loss by volatilisation of chloropicrin near the surface, especially if the surface is exposed to air currents or mechanically disturbed. Precautions based on these findings include (*inter alia*) the making of earth mounds as large as possible and keeping them covered with bitumen paper during ⩽8 days after treatment. P. S. ARUP.

**Penetration of and persistence in soil of the herbicide 3-(p-chlorophenyl)-1 : 1-dimethylurea (CMU).** L. A. Birk (*Canad. J. agric. Sci.*, 1955, **35**, 377—387).—Chemical analysis indicates that by the end of the first year approx. 90% of the CMU had disappeared; during the second year the loss averaged 62%. Using cereal crops as indicators it seems that CMU toxicity persists for relatively long periods. E. G. BRICKELL.

**Intake of solutions by tops of freshly cut oak stumps.** R. P. True, T. M. Judy and E. Ross (*Phytopathology*, 1955, **45**, 466).—The effects on the roots of oaks of several silvicides are slow and incomplete. Intakes from stump-reservoirs of solutions of azosulphamide or a water-sol. dye are considerably less, and intakes of saturated aq. $CuSO_4$ are very considerably less than intakes of water under comparable conditions. The presence of standing live companion-sprouts or other oaks root-grafted to the treated stumps increase and prolong absorption; the dye or the $CuSO_4$ is translocated to the crowns of organically connected trees. P. S. ARUP.

**Plant metabolism. V. Metabolism of radioactive 2 : 4-D in etiolated bean plants.** E. G. Jaworski, S. C. Fang and V. H. Freed (*Plant Physiol.*, 1955, **30**, 272—275).—The growth regulator is absorbed by the leaves and movement of the 2 : 4-D mol. or 2 : 4-D complex from the leaf to the stem may be induced by applying a sugar solution to the leaves, sucrose and glucose being the most effective. Radioactive 2 : 4-D is metabolised in the leaves of etiolated plants as well as in normal plants. E. G. BRICKELL.

**Tentative method for investigating the influence of application rate on the penetrative power of herbicide sprays.** P. Hebblethwaite (*J. Sci. Food Agric.*, 1955, **6**, 582—584).—A method, based on spraying with 0·5—1% solutions of Nigrosine dye, suitable for investigating penetration of herbicide spray into ground-crop foliage, is subject to a rather high sampling variation, but capable of differentiating between broadly different treatments. J. S. C.

**Effect of use of Erpan as herbicide on taste and smell of milk.** E. Flückiger (*Landw. Jahrb. Schweiz*, 1955, **69**, 357—364).—The normal use of Erpan (a compound of the 2 : 4-D type) has no effect on the taste or smell of the milk when the cows are turned out on the treated pasture after an interval of 2—3 days. Erpan is detectable by taste in concn. of 100 μg. per l. of milk. The spraying of the daily ration of a cow, 0·5 hr. before milking, with 15 g. of Erpan had no effect on the taste and smell of the milk. A reasonable interval between spraying and utilisation of the pasture is recommended as a health precaution. P. S. ARUP.

**Weeds in rice : their control in Suriname.** J. G. P. Dirven and H. J. Poerink (*Trop. Agriculture, Trin.*, 1955, **32**, 115—123).—Control measures recommended included sowing rice in a layer of water (10—15 cm.), use of clean seed, and application of "brush-killers" (2 : 4 : 5-T, MCPA and/or 2 : 4-D) 5—8 weeks after the paddy has appeared. A. G. POLLARD.

**Chemical weed-control of maize crops used to complement agricultural methods.** M. A. Maupas (*C. R. Acad. Agric. Fr.*, 1955, **41**, 520—525).—Trials are reported using ammonium dinitrophenolate and compounds of 2 : 4-D in the control of common weeds in maize crops. The best results are obtained using $NH_4$ dinitrophenolate solution 5 kg. in 700 l. of water per ha. and the triethanolamine salt of 2 : 4-D 600 g. in 200 l. of water per ha. The relative merits of the two herbicides are discussed. N. M. WALLER.

**Activities and residues of sulphur-35-labelled bis(ethyl xanthic) disulphide [Herbisan].** R. Langston (*J. agric. Food Chem.*, 1955, **3**, 849—851).—Comparative herbicidal tests indicated that the labelled laboratory-synthesised and commercial Herbisan produced an approx. 85% kill of broad-leaved weeds and 75% kill of grasses. The bis(ethyl xanthic)disulphide as formulated, was not absorbed by the vegetables tested when applied (2 gal. in a 100 gal. of water per acre) as pre-emergence spray on muck soil; none of the plants indicated a measurable quantity of radioactivity. After foliar applications to 16 vegetable crops, only in cabbage and cauliflower was there any degree of translocation as determined by autoradiographic procedures. Data indicate that bis(ethyl xanthic)disulphide may be used safely on the crops tested in regard to residue hazards. E. M. J.

**Enhancement of herbicidal activity by previous damage to weeds, using *Colchicum autumnale* as test plant.** B. Rademacher (*Z. PflKrankh.*, 1955, **62**, 605—611).—Enhanced activity of 2 : 4-D caused by previous treading-down of the plants is demonstrated by long-term field experiments. P. S. ARUP.

**Effects of rhizomes of quack (couch) grass (*Agropyron repens*) and shading on the development of weedy species.** K. C. Hamilton and K. P. Buchholtz (*Ecology*, 1955, **36**, 304—307).—The establishment of seedlings of *Veronica, Polygonum, Oxalis, Setaria* and *Trifolium* was adversely affected and of *Taxaracum and Plantago* favoured by the presence in the soil of living couch grass rhizomes. Shading favoured the establishment of all these species except *Plantago*. L. G. G. WARNE.

**Control of groundsel bush.** J. Arthur (*Agric. Gaz. N.S.W.*, 1955, **66**, 74).—Groundsel bush is controlled with one spraying of 0·2% 2 : 4-D in the early stages of growth. Older bushes are cut off and the butts painted with 1% 2 : 4-D. A. H. CORNFIELD.

## Animal Husbandry

**Comparison of peroxide- and oxygen-bomb calorimetry of feedstuffs.** J. M. Bell (*Canad. J. agric. Sci.*, 1955, **35**, 366—370).—A

tendency to high values is reported for the Parr peroxide calorimeter and is attributed to correction factors and the modern use of accelerators. The regression equation $Y = 8{\cdot}025 - 0{\cdot}2188X + 1{\cdot}7753$, where $Y$ = % error compared to true value and $X$ = % ash, permits a more accurate calculation of peroxide calorimeter data. E. G. BRICKELL.

**Relations of some biologically important amino-acids during biological decomposition in various plants.** G. Michael and B. Blume (*Arch. Tierernähr.*, 1955, **5**, 41—51).—After damp and dark storage of green lucerne, barley, clover or tobacco during four days (simulating the conditions of haymaking), the contents of free tryptophan and arginine in the sol. fraction increased by ~100—200%, whilst that of lysine increased but slightly. The extent of protein hydrolysis was considerably greater in barley and lucerne than in clover or tobacco. With respect to the total amounts of combined amino-acids originally present, losses during storage of tryptophan, arginine, and lysine were, respectively, slight, ~13%, and 30%. (43 references.) P. S. ARUP.

**Determination of crude fibre [in feeding stuffs]. III. IV.** V. P. Hirsjärvi (*Z. anal. Chem.*, 1955, **147**, 81—86, 86—99).—III. The method of V. P. Hirsjärvi and L. Andersen (cf. J.S.F.A. Abstr., 1954, ii, 27) for the determination of crude fibre is not suitable for hay or feed yeast. This method, and that of Puranen and Tomula, give lower results than the Weender method, which is recommended for such materials.

IV. Several methods for determining crude fibre in feeding stuffs containing large amounts of Ca compounds are examined, results obtained by the Weender method before incorporation of the Ca compounds being used as a basis for comparison. The methods of Mach and Lepper (*Landw. Versuchsw.*, 1926, **104**, 313) and V. P. Hirsjärvi and L. Andersen (cf. Pt. III) give satisfactory results. E. HAYES.

**Plane of nutrition and starch equivalents.** K. L. Blaxter and N. McC. Graham (*J. agric. Sci.*, 1955, **46**, 292—306).—Energy losses in sheep reached reasonably stable values after 72 hr. fasting and again 10 days after re-alimentation. The relationship between losses and food intake in terms of metabolisable energy is best described as exponential. The application and adaptation of such a relationship to cover certain phenomena in animal nutrition and to serve as a basis of prediction of energy requirements are discussed. Energy losses in excreta and body heat increase whereas the loss as $CH_4$ diminishes with rising levels of nutrition. The prediction of net energy values of feeding stuffs by conventional methods may be misleading in some cases. A. G. POLLARD.

**Nutritive effect of methionine homologues.** I. Terajima, Y. Kato and H. Ariyama (*Tohoku J. agric. Res.*, 1954, **5**, 153—158).—When added to a diet based on casein, ethionine, propionine and butionine depressed the growth of rats, ethionine producing much the greater effect. A. G. POLLARD.

**Fat in the diet increases protein efficiency.** B. Sure (*Arkansas Farm Res.*, 1955, **4**, No. 2).—In feeding trials with rats increasing proportions of fat (7—30%) in the diet resulted in an increase in the protein efficiency ratio from 0·76 to 1·0. A. G. POLLARD.

***In vitro* studies with rumen micro-organisms using carbon-14-labelled casein, glutamic acid, leucine and carbonate.** K. K. Otagaki, A. L. Black, H. Goss and M. Kleiber (*J. agric Food Chem.*, 1955, **3**, 948—951).—Rumen fluids withdrawn from mature wethers and incubated with carbon-14-labelled casein, 1-carbon-14-glutamic acid, leucine-3-carbon-14, and $^{14}CO_2$ indicated that proteolytic enzymes are active in the rumen fluid, and that deaminases or transaminases decompose the amino-acids giving volatile fatty acids and $CO_2$. The micro-organisms also fix C from $CO_2$ in both essential and non-essential amino-acids. (19 references.) E. M. J.

**Feeding of livestock.** J. Duckworth (*J. roy. agric. Soc.*, 1954, **115**, 174—183).—A review dealing principally with the use of vitamins (notably E and $B_{12}$) and of fish and whale solubles in stock feeds. A. G. POLLARD.

**Vitamin K and mammary gland development.** B. Dodi (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 20—26).—Vitamin K (2-methyl-1 : 4-naphthaquinone) in large doses (up to 135 mg.) exerts in guinea-pigs a marked plastic effect on the mammary gland, even in castrated animals. F. R. PAULSEN.

**Feeding value of sweet potato tubers.** M. H. French (*E. Afr. agric. J.*, 1955, **21**, 18—19).—Chemical composition, digestibility data, and starch equiv. of sweet potato tubers are presented. The tubers are largely carbohydrate and have a high digestibility. A. H. CORNFIELD.

**Potato ensilage.** F. Hansen (*Tidsskr. Planteavl.*, 1955, **59**, 196—215).—Hot cooked potatoes were placed directly into concrete silos and mashed. The loss of dry matter during ensilage averaged 6—10% in 4—7 months. Alternatively raw potatoes, whole, grated or frozen, were packed into a silo, compressed and covered with paper and a layer of 30—50 cm. of sand. The $O_2$ content of the atm. in the silo decreased to zero and the $CO_2$ content rose to 21%. Subsequently the $CO_2$ content increased further to 40—80% as the substance of the tubers collapsed: a considerable amount of drainage liquor separated. The total loss of wt. under these conditions approached 50%, the finished silage containing 35—40% of dry matter. The dry matter of the drainage liquor, representing 1·5—6·0% of the initial dry matter of the tubers, contained nearly half its wt. of org. N substances. Net losses of dry matter during ensilage of cooked, uncooked, whole, grated and frozen tubers were 8·3, 15·0, 8·5 and 12·0% respectively. Addition of $H_2SO_4$ to the uncooked grated potatoes lowered the loss of dry matter in the silo to 9%. A. G. POLLARD.

**Necessity for checking oxytocic power of fish-meals.** B. Dodi (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser ii, 13—19).—Testing of fish-meals on rat uterus *in vitro* is advisable if the meals are for animal feeding, as poor quality meals contain an oxytocic principle which has caused abortion in cows. F. R. PAULSEN.

**Cottonseed meal in poultry feed: inactivation of gossypol by treatment with phloroglucinol.** C. R. Grau, T. L. Lau and C. L. Woronick (*J. agric. Food Chem.*, 1955, **3**, 864—865).—Cottonseed meal treated with phloroglucinol gives a product that contains no detectable available gossypol, as measured by the gossypol–cephalin contents of eggs from hens fed the meal. Further data are needed before the process can be used in the manufacture of cottonseed meals for poultry feeding. E. M. J.

**Application of a Latin square change-over design to dairy cattle grazing experiments.** N. R. Thompson, R. E. Blaser, G. C. Graf and C. Y. Kramer (*J. Dairy Sci.*, 1955, **38**, 991—996).—This design is more efficient than single grouping or complete randomisation, but requires more work to analyse the data and may lead to some confounding of effects of pasture mixture with effects of other variables. S. C. JOLLY.

**Effects of the maize–lucerne hay ratio on the digestibility of different nutrients by cattle.** T. W. Dowe, J. Matsushima and V. Arthaud (*J. Anim. Sci.*, 1955, **14**, 340—349).—In a series of rations of similar crude and digestible protein contents the ratio of maize : lucerne hay was varied from 1 : 1 to 5 : 1. Increases in the proportion of maize were associated with increase in the dry matter, crude protein and N-free extractives in the fæces and in the apparent digestibility and ether extract of the ration. The coeff. of apparent digestibility of the N-free extract, crude fibre and protein were unaffected. A. G. POLLARD.

**Nutrient element content of native forages in relation to location and land forms in the South Carolina coastal plain.** K. C. Beeson (*Soil Sci.*, 1955, **80**, 211—220).—Two species of the native vegetation were sampled in different areas, defined according to their geological characteristics. A terrace with an elevation of 25 to 42 feet above sea level was associated with low levels of the nutrients, whilst a higher terrace was associated with a higher mineral content in the vegetation. Drainage basins produced vegetation low in P, Co, Cu and Fe but high in Ca and Mn. Co levels were generally low. T. G. MORRIS.

**Influence of high sodium chloride intakes by fattening sheep and cattle.** J. H. Meyer, W. C. Weir, N. R. Ittner and J. D. Smith (*J. Anim. Sci.*, 1955, **14**, 412—418).—When added to sheep rations at varying rates, up to 12·8% (0·46 lb. daily intake per head) salt had no detrimental effects on the digestibility of the total ration or of the crude protein, on the retention of N, on the daily gain in wt. or on the feed efficiency. With daily intakes of 0·33 and 0·4 lb. of salt the wt. of the kidneys increased but the wt. of the adrenals was unchanged: blood-albumin and hæmatocrit values were unaffected. Carcass quality was affected only by the 0·4 lb. daily dosage. The no. of stomach worms was unchanged. High-salt rations given to steers (up to 1·7 lb. per head daily) had no ill effects on growth rates or feed efficiency. The carcass dressing (%) was unchanged and carcass quality was adversely influenced only by the highest proportion of salt used (1·7 lb.). A. G. POLLARD.

**Availability of iodate iodine to sheep.** Evan Wright and E. D. Andrews (*N.Z. J. Sci. Tech.*, 1955, **37**, 83—87).—Oral administration of radioactive $^{131}I$ (as iodate and as iodide) to six sheep indicated that, in four of the animals, comparable amounts of $^{131}I$ were accumulated by thyroid glands from iodate and from iodide. In the other two animals, uptake from iodate was much lower than from iodide. In all sheep, however, the amounts of iodate-I accumulated showed that $KIO_3$ is a suitable source of I for sheep. (16 references.) R. H. HURST.

**Apparatus for measuring motility of sperm cells.** C. A. Bosselaar, N. Spronk and G. C. van Dam (*J. agric. Sci.*, 1955, **46,** 417—419).—Using apparatus previously described (Bosselaar and Spronk, *Nature, Lond.*, 1952, **169,** 18), the image of the moving sperm is transmitted to a photomultiplier. Voltage pulsations resulting from the movement of the sperm cells are amplified, passed through a discriminator and counted. Measurement of the concn. of sperm cells on the slide presents some difficulty and limits the accuracy of determinations of motility. A. G. POLLARD.

**Effect on conception rates of semen diluents containing citrate or phosphate buffer, with all combinations of sulphanilamide, streptomycin and penicillin.** R. C. Campbell and J. Edwards (*J. agric. Sci.*, 1955, **46,** 44—55).—Results of a factorial experiment involving 68,713 first inseminations at four centres is reported. When unsupplemented, the phosphate buffer gave a conception rate 5·5% greater than did the citrate buffer. The citrate buffer with sulphanilamide, streptomycin and penicillin gave a conception rate 6·8% above that obtained with unsupplemented citrate buffer. Phosphate buffer with penicillin gave a conception rate 9·3% above that obtained with unsupplemented citrate buffer. Sulphanilamide or streptomycin depressed the conception rate of phosphate buffer. A. H. CORNFIELD.

**Habits of Zebu cattle. III. Water consumption.** D. H. L. Rollinson, K. W. Harker and J. I. Taylor (*J. agric. Sci.*, 1955, **46,** 123—129).—Water consumption and its relationship to temp., R.H. and extent of grazing are reported. A. H. CORNFIELD.

**Indole content of the rumen of cattle.** D. Spisni and V. Cappa (*Ann. Fac. Agr. Univ. cattol. S. Cuore,* 1955, **51,** Ser. i, 392—396).—The large urinary excretion of indican by cattle is probably due to presence of indole in the rumen, to the extent of ~0·73 mg. per l. of filtrate. F. R. PAULSEN.

**Inflatable urethral catheter for urine collection from cows.** H. M. Cunningham, G. L. Frederick and G. J. Brisson (*J. Dairy Sci.*, 1955, **38,** 997—999).—The urethral catheter described is of inflatable rubber and enables collection of urine from dairy cows and heifers for periods up to three weeks without causing infection or imposing undue strain on the animal. S. C. JOLLY.

**Effect of feeding on indoxyl compounds in urine of cows.** D. Spisni and V. Cappa (*Ann. Fac. Agr. Univ. cattol. S. Cuore,* 1955. **51,** Ser. i, 380—391).—In cows, change of diet from hay to meadow grasses caused a sharp decrease in urinary indoxyl-compounds, while a change to a leguminous diet had the opposite effect. F. R. PAULSEN.

**Yields of holocellulose prepared from ruminant fæces by acid chlorite treatment.** R. E. Ely and L. A. Moore (*J. Dairy Sci.*, 1955, **38,** 1017—1022).—Satisfactory recoveries of holocellulose (**I**) were obtained by the method of Ely *et al.* (cf. J.S.F.A. Abstr., 1954, ii, 287) from 5 fæces samples from cows fed 10 different forages as their entire ration; the remaining five recoveries were higher than the theoretical **I** contents due to a noncarbohydrate or non-reducing-carbohydrate constituent. Repeated acid chlorite treatment caused loss of carbohydrate from **I** fractions with certain samples, so that theoretical yields of **I** cannot be obtained from all fæces by a fixed no. of acid chlorite treatments. S. C. JOLLY.

**Effect of various restricted diets on growth and on certain blood components of young dairy calves.** J. M. Wing, N. L. Jacobson and R. S. Allen (*J. Dairy Sci.*, 1955, **38,** 1006—1016).—Plasma-vitamin A levels of calves on a diet containing milk fat (~18,000 i.u. of total vitamin A equiv. per 100 lb. of body wt. daily) from 4 to 60 days of age were maintained, but those of animals on low-fat (*A*) or hydrogenated-soya-bean-oil (*B*) diets decreased despite supplementation with 12,000 i.u. of vitamin A per 100 lb. of body wt. daily. Mean plasma-fat concn. were highest in animals fed whole milk, followed in order by those in animals fed butter oil, *B*, and *A*. Mean blood-reducing-sugar levels decreased in all groups, but generally were highest in animals fed *A* (high in lactose). Wt. gains and plasma-Ca, -inorg. P, and -hæmoglobin were similar in all groups. S. C. JOLLY.

**Effect of glycuronic lactone on ascorbic acid content of blood, urine and milk of cows.** V. Cappa (*Ann. Fac. Agr. Univ. cattol. S. Cuore,* 1955, **51,** Ser. i, 373—379).—In cows intravenous injection of glycuronic acid γ-lactone produces no increase in ascorbic acid in blood plasma, milk or urine. F. R. PAULSEN.

**Effect of oral administration of hormones on growth rate and deposition in the carcass of fattening steers.** T. W. Perry, W. M. Beeson, F. N. Andrews and M. Stob (*J. Anim. Sci.*, 1955, **14,** 329—335).—Diethylstilbœstrol or hexœstrol, fed to 2-year steers at the rate of 10 mg. daily, significantly increased the gain in live wt. Dienœstrol produced similar though slightly smaller effects. The œstrogens lowered food consumption per unit increase in wt. and induced certain pelvic and mammary modifications. Carcass grading after administration of stilbœstrol was slightly lower than that in controls or in animals receiving the other œstrogens. There was no residue of œstrogen in the meat whether the treatment was discontinued one or seven days prior to slaughter. A. G. POLLARD.

**Aureomycin effects: growth and digestibility studies with identical twin calves.** G. I. Pritchard, J. A. Newlander and W. H. Riddell (*J. Anim. Sci.*, 1955, **14,** 336—339).—Oral administration of aureomycin to calves accelerated growth, lowered food consumption per unit gain in wt. but did not affect the digestibility of dry matter, ash, protein, crude fibre, N-free extractives or fat. A. G. POLLARD.

**Effects of feeding different grades of hay and cod-liver oil concentrate to dairy cattle. III. From 361 to 720 days of age.** H. B. Ellenberger, J. A. Newlander and C. H. Jones (*Vermont agric. Exp. Sta.*, 1954, *Bull.* 576, 44 pp.).—On the whole yearling heifers did as well on poor-quality hay (mostly timothy, cut late July to early Aug.) as on good-quality hay (cut late June to early July) when these were supplemented with succulent roughages and grain. Vitamins A and D supplement was of some value in improving the feeding quality of the poor-, but not of the good-quality, hay. A. H. CORNFIELD.

**Cattle fattening on permanent grass and leys.** T. E. Williams and W. Davies (*J. roy. agric. Soc.*, 1954, **115,** 98—111).—In grazing experiments with sheep and cattle comparison is made of permanent pastures and leys at three centres. Live-wt. yields per acre varied with the nature and manurial treatment of the soil, with the dominant plant species and with grazing management. Deterioration of ley swards up to five years was negligible provided suitable manuring and stocking was practised. A. G. POLLARD.

**Comparison of high-quality and low-quality hay for raising dairy calves (birth to 24 months of age).** J. A. Newlander and W. H. Riddell (*Vermont agric. Exp. Sta.*, 1954, *Bull.* 577, 8 pp.).—The high-quality hay was early-cut mixed hay, mostly clover and lucerne with some bromegrass and timothy. The low-quality hay was from old meadow consisting largely of Kentucky bluegrass, redtop, quack and timothy. The former type was higher in protein and lower in fibre. When these rations were supplemented with limited amounts of grain, the group on high-quality hay made 10·2% greater wt. gains than did the group on low-quality hay. A. H. CORNFIELD.

**Relation between feeding and milk production in cows.** G. Piana and F. Usuelli (*Ann. Fac. Agr. Univ. cattol. S. Cuore,* 1955, **51,** Ser. i, 256—275).—In ruminants the milk-fat is synthesised from acetic acid derived from the breakdown of carbohydrates by micro-organisms in the rumen. Feeding has a marked effect on the organoleptic qualities of milk and the butter produced from it and on the m.p. of the butter. This last property is lowered by feeding red clover, linseed cake, colza, sesame, soya, sunflower, maize, oat and wheat-bran, but raised by potato, beet palm-cake and cotton-cake, seeds of leguminosæ, rye, barley and peas. Excessive cattle-cake leads to a cream which froths excessively in the churn, making butter preparation impossible. F. R. PAULSEN.

**Methods of milk production; survey in four areas of England and Wales.** F. B. Leech, J. W. Edgell, P. Heskin and S. B. Thomas (*J. agric. Sci.*, 1955, **46,** 78—89).—Methods, buildings and equipment used for milk production in four counties are reported from a random sample survey carried out in 1948—9. A. H. CORNFIELD.

**Milk composition studies in the hormonal induction of lactation using identical twin dairy cattle.** D. R. Perrin (*N.Z. J. Sci. Tech.*, 1955, **37,** 88—92).—The heifers were brought into lactation by treatment with progesterone and œstrogen. In general, the mammary secretions were initially colostral in nature, but gradually altered to a milk with normal contents of fat, protein, lactose and mineral constituents. R. H. HURST.

**Feeding value of excellent forage for milk production.** M. E. McCullough and O. E. Sell (*J. Dairy Sci.*, 1955, **38,** 1023—1027).—Milk production of >40 lb. daily was maintained on winter forage consisting of oats, rye-grass and crimson clover, so long as the dry-matter digestibility was >70%. A 20-lb. level of production was maintained on free-choice feeding of U.S. No. 2 hay supplemented with a daily 2-hr. grazing of the forage. A 28-lb. level of production was maintained when the 2-hr. grazing supplemented good maize silage and U.S. No. 1 lespedeza hay. The forage could not maintain production when fed with poor hay. Free-choice feeding of good roughage may reduce the amount of pasture consumed and thus increase the no. of cows that can be grazed per acre. S. C. JOLLY.

**Effect of frequency of ejaculation on semen production, seminal characteristics and libido of bulls during the first post-pubertal year.** F. N. Baker, N. L. VanDemark and G. W. Salisbury (*J. Dairy Sci.*, 1955, **38,** 1000—1005).—Bulls might be placed in service younger and used more frequently than hitherto recommended. Overt

sexual interest was first expressed at ~29 weeks and ability to ejaculate first demonstrated at ~38 weeks of age. Service ⩽3 times a week does not apparently affect adversely seminal characteristics or spermatozoa production except in so far as the latter is affected by reduced libido. S. C. JOLLY.

**Metabolism of bull semen. I. Inorganic and total phosphorus relations.** F. H. Flerchinger and R. E. Erb (*J. Dairy Sci.*, 1955, **38**, 1028—1036).—Total P was markedly related to spermatozoa concn. (*C*) in semen from high-fertility bulls. Inorg. P, both before and after 1-hr. incubation at 37°, was most closely correlated with *C*, but little related to initial mobility, fructose and fructose utilisation. During the incubation inorg. P increased by 1·4 mg. per 100 g. (8·2%); the change was not related to bulls, breeds, months or any semen quality measured. Inorg. and total P were not related to non-return rate. S. C. JOLLY.

**Influence of vitamins and hormones on vitality of stored bull semen.** G. M. Curto (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 401—403).—Lutein and deoxycorticosterone acetate in oily solutions increase the survival of the spermatozoa, but in aq. solution they depress the vitality. Thiamine and ascorbic acid have no effect. F. R. PAULSEN.

**Effect of heated fortified skim milks on the livability of bovine spermatozoa.** R. G. Saacke, J. O. Almquist and S. Patton (*J. Dairy Sci.*, 1955, **38**, 1046—1047).—Fresh skim milk fortified by the addition of condensed skim milk, superheated condensed skim milk, or skim milk powder is unsuitable as a diluent for bovine semen unless diluted with water to reduce the content of non-fat solids to that of normal skim milk. S. C. JOLLY.

**Feeding lambs on pasture and supplementation of ewes with phenothiazine at breeding time.** G. W. Litton, C. M. Kincaid and R. E. Hunt (*Virginia agric. Exp. Sta.*, 1954, *Bull.* 468, 14 pp.).—When lambs on good pasture were supplied with supplemental grain there was no significant improvement in wt. gains or carcass grade, but dressing % was increased slightly. Drenching ewes during the breeding season with 50 g. of phenothiazine every week for eight weeks had no significant effect on conception. A. H. CORNFIELD.

**Protein requirements of fattening feeder lambs.** L. F. Bush, J. P. Willman and F. B. Morrison (*J. Anim. Sci.*, 1955, **14**, 465—469).—Lambs were fed rations containing 10, 11 or 11·8% of protein supplemented with (*a*) maize silage as sole roughage, (*b*) 0·75 lb. of lucerne hay with silage or (*c*) 0·50 lb. of the hay with silage, daily. The higher protein rations produced the more rapid gain in wt. but the lambs were less fat. Differences in protein levels did not affect the feed efficiency of the ration. Maize silage as sole roughage produced more efficient and more economical increases in wt. than when used with lucerne hay but silage with the smaller proportion of lucerne hay gave a somewhat better finish. A. G. POLLARD.

**Milk yields of Scottish Blackface ewes.** J. Munro (*J. agric. Sci.*, 1955, **46**, 131—136).—At two locations the estimated milk yields of Blackface ewes with single lambs ranged from 65 to 220 lb. over six weeks. Daily live-wt. increases of lambs ranged from 0·23 to 0·91 lb. At one location the average lactation curve reached a max. 24 days after lambing, whilst, at the other, max. occurred 10 and 32 days after lambing. An average of 5·5 oz. milk was required to give 1 oz. live-wt. increase in lambs. The chemical composition of the milk is given. A. H. CORNFIELD.

**Response of cobalt-deficient lambs to orally administered vitamin $B_{12}$.** C. J. Kercher and S. E. Smith (*J. Anim. Sci.*, 1955, **14**, 458—464).—In respect of appetite, body wt. and hæmoglobin levels, Co-deficient sheep responded equally to vitamin $B_{12}$ whether orally administered at the rate of 500 $\mu$g. daily for five weeks or injected (500 $\mu$g.) over a period of 14 days. A. G. POLLARD.

**Effect of cobalt and calcium on synthesis of vitamin $B_{12}$ in sheep raised in cobalt-deficient biogeochemical areas.** V. V. Koval'skii and Yu. I. Rayetskaya (*Dokl. Akad. Nauk SSSR*, 1955, **100**, 1131—1134).—In sheep with acobaltosis only traces of vitamin $B_{12}$ are found in the liver, the Co content of which is not, however, lower than in healthy animals receiving the same diet. The vitamin $B_{12}$ content is doubled by adding 2 mg. of $CoCl_2$ to the daily ration, and still further raised by adding chalk to the diet. Where the soil is also deficient in Cu and I, these elements should also be added to the ration, to assure vitamin $B_{12}$ synthesis. R. TRUSCOE.

**The skeleton of the sheep. I. Effect of different levels of dietary calcium during pregnancy and lactation on individual bones.** D. Benzie, A. W. Boyne, A. C. Dalgarno, J. Duckworth, R. Hill and D. M. Walker (*J. agric. Sci.*, 1955, **46**, 425—440).—The level of Ca intake (1·4, 4·5 or 7·4 g. daily) by ewes during pregnancy did not affect the no. of lambs born, their wt. at birth or rate of growth during suckling. The smallest Ca intake induced the lowest blood-Ca level and the greatest resorption of bone substance: blood-org. P was unaffected by the Ca intake. Resorption of Ca during lactation on a Ca-deficient diet was greatest in the vertebræ and pelvis and least in the shafts of long bones. The wt. of the ash of bones was a better measure of resorption than was the % of ash. In general, radiological examination of bones confirmed the results of chemical analysis in many but not in all respects. A. G. POLLARD.

**Thyroid secretion rate of sheep as affected by season, age, breed, pregnancy and lactation.** H. A. Henneman, E. P. Reineke and S. A. Griffin (*J. Anim. Sci.*, 1955, **14**, 419—434).—Using injected $^{131}I$ thyroid secretion rates in sheep are shown to vary seasonally (min. in July) and to increase during lactation but not during pregnancy. Differences due to breed were apparent only in Dec. and Jan. A. G. POLLARD.

**Effect of testosterone and pregnant mare serum on semen production of rams of low libido.** S. I. Ahmed (*J. agric. Sci.*, 1955, **46**, 168—172).—The libido of initially sexually inactive rams was increased to a greater extent by implanting subcutaneously a 0·1 g. pellet of testosterone propionate than by weekly injections with 2000 i.u. of pregnant mare serum. The testosterone treatment maintained high libido even during summer whilst that of the sexually active control rams usually fell off during this period. The testosterone treatment increased the vol. of ejaculate, and % of dead spermatozoa and reduced the impedance-change frequency. Motility ratings were unaffected by any of the treatments. A. H. CORNFIELD.

**Effect of glycine on storage of ram semen.** S. I. Ahmed (*J. agric. Sci.*, 1955, **46**, 164—167).—Replacing the citrate buffer in a standard citrate–egg-yolk diluent with 4% glycine considerably improved the survival of ram semen stored in the buffer at 4°. A semen dilution ratio of 1 : 20 gave better survival than did a ratio of 1 : 40 when the 4% glycine–egg-yolk buffer was used. Results were similar whether 33·3% or 50% of egg yolk was present in the buffer, whilst with 25% egg yolk there was a slight reduction in motility. A. H. CORNFIELD.

**Use of sheep in controlling skeleton weed.** E. Tindale (*Agric. Gaz. N.S.W.*, 1955, **66**, 369—371).—A review and general discussion. A. H. CORNFIELD.

**Electrically warmed floors for fattening pigs.** R. S. Barber, R. Braude and K. G. Mitchell (*J. agric. Sci.*, 1955, **46**, 31—36).—Heating the pen floors by two electrical methods over 8—15 weeks had no significant effect on wt. gains or feed efficiency of fattening pigs. A. H. CORNFIELD.

**Interaction between environment and level of feeding for pigs from weaning to bacon weight.** I. A. M. Lucas and A. F. C. Calder (*J. agric. Sci.*, 1955, **46**, 56—77).—With a high plane of feeding (level of energy nutrition) there was little difference in wt. gains or feed efficiency from 45 to 100 lb. live-wt. between pigs kept in a good piggery (insulated building with outside run) and those kept in a bad piggery (relatively cold building with no outside run) either during winter or summer. With a low plane of feeding there was no difference in results between winter and summer in a good piggery, whilst results were much poorer in winter than in summer in a bad piggery. Efficiency of conversion of total digestible nutrients to live-wt. gains from 45 to 100 lb. was better with well-housed pigs at a low plane of feeding than with pigs in either type of house at a high plane of feeding. During the growth of pigs from 100 to 200 lb. live-wt. the feed efficiency of well-housed low-plane pigs was better than that of well-housed high-plane pigs but was not better than that of high-plane poorly-housed pigs. A. H. CORNFIELD.

**Influence of crude fibre content of fodder on growth of young pigs.** J. Axelson (*Arch. Tierernähr.*, 1955, **5**, 1—16).—In comparative feeding trials, increases in the crude fibre content (4·8—9·3) decreased (linearly) the digestibility of the components of the feed; these effects decreased with the growth of the pigs (30—90 kg.). Throughout the experiment, the optimum crude fibre level was 6·6% with respect to gain in wt., and 7·2 with respect to energy yield. Increases in crude fibre had no adverse effect as regards transport and slaughtering losses, or the quality of the carcasses. The results are discussed in relation to feed evaluation. P. S. ARUP.

**Histidine: an essential nutrient for the growth of pigs.** R. G. Eggert, L. A. Maynard, B. E. Sheffy and H. H. Williams (*J. Anim. Sci.*, 1955, **14**, 556—561).—Experimental evidence presented demonstrates the need of dietary histidine for young growing pigs. A. G. POLLARD.

**Some effects of adding supplements of lysine, methionine and tryptophan to practical swine rations.** W. H. Pfander and L. F. Tribble (*J. Anim. Sci.*, 1955, **14**, 545—555).—L-Lysine (0·1%), added to a ration comprising maize, soya-bean meal, tankage and wheat shorts, increased the growth rate of the pigs and the nutrient

efficiency of the ration without affecting food consumption appreciably. Methionine and tryptophan produced similar but smaller effects whether given separately or in combination. Addition of lysine and methionine, separately or together, to a maize–soya-bean meal ration increased growth rates without affecting the feed efficiency. The amino-acid requirements of pigs are represented as, L-lysine 5·0, methionine 3·5 (with cystine) and tryptophan 1·0% of the protein. A. G. POLLARD.

**Utilisation of lard by baby pigs.** H. M. Cunningham and G. J. Brisson (*Canad. J. agric. Sci.*, 1955, **35**, 371—376).—The level of lard in the diet fed to 2-day-old baby pigs had no effect on the apparent digestibility of fat, casein or glucose, or on the efficiency of energy utilisation, but the relationship between true and apparent digestibility was affected, the former being somewhat higher. E. G. BRICKELL.

**Effects of various sequences of full and limited feeding on the reproductive phenomena in Chester White and Poland China gilts.** H. L. Self, R. H. Grummer and L. E. Casida (*J. Anim. Sci.*, 1955, **14**, 573—592).—Effects of different alternations of feeding levels (full and $\frac{2}{3}$-full rations), on ovulation rate, embryo survival, age at puberty and rate of gain in wt. immediately before and after the breeding season are examined. No significant differences between breeds in these effects were apparent. A. G. POLLARD.

**Milk production of large white pigs.** R. S. Barber, R. Braude and K. G. Mitchell (*J. agric. Sci.*, 1955, **46**, 97—118).—Under natural conditions the average interval between successive sucklings was about 1—1·25 hr. When suckling was allowed every hour the quantity of milk obtained, live-wt. gains and efficiency of utilisation of milk was better than when suckling was allowed only every 2·5—3 hr. The estimated average 56-day lactation yield was 768 lb. (range 655—882 lb.), although the true yield may be 5—10% higher. Live-wt. gains of pigs were closely related to milk intake during the first three weeks of life but not during the last five weeks of lactation, during which supplementary food was available. The supply of sow's milk may frequently be insufficient for optimum growth of pigs. Data are also given on the effect of light on milk yields and chemical composition of sow's milk in relation to stage of lactation. A. H. CORNFIELD.

**Growth of pigs kept to one level of feeding, in two environments, and fed diets with and without an antibiotic.** I. A. M. Lucas and A. F. C. Calder (*J. agric. Sci.*, 1955, **46**, 307—319).—Pigs growing under " bad " (colder, more damp and draughty) and those under " good " piggery conditions showed no significant differences in growth rate (up to 100 lb. live wt.) or feed efficiency. During the finishing period (100—200 lb.) differences in growth rate were insignificant but feed efficiency was greater under " bad " conditions. " Good " conditions resulted in fatter carcasses with higher killing-out %. Supplementary feeding of procaine penicillin (12 g. per ton of feed) did not affect growth rates or carcass measurements but caused a larger killing-out %. A. G. POLLARD.

**Protein levels for pigs as studied by growth and self selection.** J. W. Lassiter, S. W. Terrill, D. E. Becker and H. W. Norton (*J. Anim. Sci.*, 1955, **14**, 482—491).—The min. protein requirement of growing pigs varied somewhat with the nature of the ration and with the method of feeding. In dry-feed experiments described, using a ration based on maize–protein supplement, the min. protein requirement was 14—16 % up to 100 lb. live-wt. If the same protein level is maintained for feeding up to 200 lb. live-wt. the min. protein level was 12—14%. For pigs on lucerne pasture the min. level was 12—14% up to 200 lb. live-wt. with a preference for the 14% level from weaning to 100 lb. live-wt. Pastured pigs given free choice of maize and protein supplement selected 10·8% of protein, up to 100 lb. live-wt. and 10% thereafter. A. G. POLLARD.

**Influence of arsanilic acid on protein requirements of growing pigs.** L. E. Hanson, E. F. Ferrin and S. N. Singh (*J. Anim. Sci.*, 1955, **14**, 525—531).—The growth of pigs on rations of different protein contents (14—18% from weaning to 125 lb. live-wt. and 11—15% from 125 to 200 lb. live-wt. with and without supplements of arsanilic acid) is recorded. The higher protein levels produced more rapid growth in the early stages but the marketing wt. was reached in substantially the same time with all dietary protein levels used. Arsanilic acid (60 g. per ton of feed or 240 g. per ton of protein supplement given " free-choice " with maize meal) did not produce significant increases in live-wt. but improved feed efficiency by about 4% : it had no protein-sparing effect and no toxic action. A. G. POLLARD.

**Use of arsanilic acid in the production of market pigs.** L. E. Hanson, L. E. Carpenter, W. J. Aunan and E. F. Ferrin (*J. Anim. Sci.*, 1955, **14**, 513—524).—Arsanilic acid (30—90 g. per ton of feed) in pig rations slightly increased the average daily gain in wt. but not the feed efficiency. Carcass quality was unaffected. Small amounts of As accumulated in the tissues. On cessation of feeding arsanilic acid a few days before slaughter As was eliminated rapidly from liver and kidneys but more slowly from muscle. A. G. POLLARD.

**Riboflavin studies with pigs.** S. W. Terrill, C. B. Ammerman, D. E. Walker, R. M. Edwards, H. W. Norton and D. E. Becker (*J. Anim. Sci.*, 1955, **14**, 593—603).—Pigs receiving a purified diet *ad lib.* were subjected to a preliminary riboflavin-depletion period and were then given various levels of riboflavin (**I**). The final wt. of the pigs and their feed consumption were similar for rations containing $<0{\cdot}65$ mg. of **I** per lb. of ration, but were smaller for lower levels of **I**. Neither vitamin $B_{12}$ (15 $\mu$g.) nor chlortetracycline (10 mg. per lb. of ration) had any significant influence on the **I** requirement of the pigs. Beneficial effects of **I** on growth rates were largely accounted for by its action in increasing the food intake : the optimum level was 0·4—0·65 mg. per lb. of ration with an environmental temp. of 53°F. A. G. POLLARD.

**Effects of the addition of dehydrated lucerne meal, fish solubles and a " vitamin $B_{12}$ " concentrate to a purified diet for sows.** D. I. Gard, S. W. Terrill and D. E. Becker (*J. Anim. Sci.*, 1955, **14**, 562—572).—Sows receiving a purified control diet supplemented with lucerne meal (10%) showed a greater capacity for weaning litters and subsequently required smaller average no. of services per conception than did those given the unsupplemented ration. Wt. per litter were unaffected. Sows given supplements of menhader fish solubles (3% of ration) or a " vitamin $B_{12}$ " concentrate produced heavier litters which were heavier at weaning than were control litters, but neither supplement had any beneficial influence on gestation or lactation. A. G. POLLARD.

**Sources of unidentified factors for the pig.** D. I. Gard, D. E. Becker, S. W. Terrill, H. W. Norton and A. V. Nalbandov (*J. Anim. Sci.*, 1955, **14**, 532—544).—An unidentified growth factor for pigs probably occurred in grass juice concentrate but none was evident in dried brewers' yeast, dried whey with whey fermentation solubles or menhaden fish solubles. Depression of growth due to inclusion of 10% of lucerne meal was not attributable to its fibre content. A. G. POLLARD.

**Some effects of œstrogen injections on the œstral cycle of gilts.** H. E. Kidder, L. E. Casida and R. H. Grummer (*J. Anim. Sci.*, 1955, **14**, 470—474).—Injections of 3 mg. of stilbœstrol on the eleventh day of the œstrous cycle lengthened the cycle ; those given on the 16th day had variable effects but tended generally to shorten the cycle. Those given on the sixth day had no effect. A. G. POLLARD.

**Effect of orally administered stilbœstrol and testosterone on growth and carcass composition of swine.** W. M. Beeson, F. N. Andrews, T. W. Perry and M. Stob (*J. Anim. Sci.*, 1955, **14**, 475—481).—Oral administration of methyl testosterone (20 mg.) or of diethyl stilbœstrol (2 mg. per head daily) with or without Terramycin did not improve growth rate or feed efficiency. Testosterone treatment produced carcasses having heavier lean cuts and lighter fat cuts than those from normally fed animals or those given a Terramycin supplement. A. G. POLLARD.

**Effects of antibiotics in pig-feeding.** A. Szilvinyi and H. Leithenmayr (*Mitt. VersSta. Gärungsgew.*, 1955, **9**, 125—129).—Terramycin, aureomycin, Vaupen and Mastpen have approx. the same effects as regards increased gains in wt. during a 70-day supplementation period. Changes in the fæcal flora during supplementation consist in a suppression of the Gram-positive, and a large increase in the coliform flora. Such increases due to Terramycin and aureomycin are slower but more persistent (taking into account the post-supplementation period) than those due to the other two (penicillin) supplements. Differences between the effects of the two pairs of antibiotic prep. are found with respect to other bacterial types. The observed increases in gains in wt. cease with discontinuation of supplementation. P. S. ARUP.

**Supplementary protein and the response of pigs to antibiotics.** D. E. Becker, S. W. Terrill and R. A. Notzold (*J. Anim. Sci.*, 1955, **14**, 492—498).—The rate of growth of weanling pigs on a maize–soya-bean oil-meal ration was increased by supplementary feeding of chlortetracycline (**I**) (6 mg. per lb. of ration) ; a mixture of equal parts of **I**, procaine penicillin and streptomycin produced still better responses and, if used with a maize–fish meal ration improved both growth rate and feed efficiency. The maize–fish-meal ration (tryptophan = 0·10%) without antibiotic supplement gave better results after further addition of 0·05% of DL-tryptophan. A. G. POLLARD.

**Effect of antibiotic dietary supplements on the carcass measurements and dressing percentage of bacon pigs.** G. Harrington and J. H. Taylor (*J. agric. Sci.*, 1955, **46**, 173—179).—Carcass length, back-fat measurements and belly thickness of bacon pigs were not generally significantly affected by addition of penicillin (15—28·4 g.)

or aureomycin (14·4—28·8 g.) per ton of feed to the diets. Dressing % was increased by aureomycin but not by penicillin. Animal protein diets gave higher dressing % than did vegetable protein diets. Carcass length in the penicillin-treated group showed much more variation than in the control or aureomycin-treated groups.
A. H. CORNFIELD.

**Effect of aureomycin on growth and reproduction in swine.** R. J. Davey, W. W. Green and J. W. Stevenson (*J. Anim. Sci.*, 1955, **14**, 507—512).—The average daily gain in wt. of pigs from weaning to 220 lb. live-wt. was increased by feeding aureomycin, optimum results being obtained with 50 mg. per lb. of ration; a 100-mg. dosage was less effective. Continuous administration of aureomycin during the breeding season did not affect the reproductive performance of the animals. A. G. POLLARD.

**Nutrition of the bacon pig. XVIII. Influence of dietary penicillin on the growth rate, efficiency of food conversion and nitrogen retention.** R. E. Evans (*J. agric. Sci.*, 1955, **46**, 329—361).—The principal effect of feeding procaine penicillin to young pigs was to stimulate appetite. At full growth the effects had largely disappeared and any remaining beneficial action was of no economic importance.
A. G. POLLARD.

**Effect of graded levels of lucerne and aureomycin on growing-fattening swine.** V. R. Bohman, J. E. Hunter and J. McCormick (*J. Anim. Sci.*, 1955, **14**, 499—506).—Inclusion of increasing amounts of lucerne meal (up to 50%) in a ration based on ground milo with a protein supplement of cottonseed meal, meat scrap and fish meal resulted in the production of leaner carcasses but also a diminution in rate of gain in wt. Aureomycin supplements increased the rate of growth and the gain in wt. per unit food consumed. High-level feeding of roughage induced enlargement of the digestive tract.
A. G. POLLARD.

**Effect of APF supplements on rate of growth of pigs receiving adequate protein supplies.** H. Krause and G. Vogel (*Arch. Tierernähr.*, 1955, **5**, 17—25).—Starting with pigs at 65 kg., daily supplementation with 15 g. of the prep. " Betapan " shortens the period required to reach the final wt. (120 kg.) by four weeks. The economic aspects of the procedure are favourable. P. S. ARUP.

**Digestibility of the carbohydrate complex of barley, wheat and maize by adult fowls.** W. Bolton (*J. agric. Sci.*, 1955, **46**, 119—122).—The sugar and starch in barley, wheat and maize were completely digested by adult fowls, about 33% of the pentosan was digested, whilst the cellulose and lignin were not digested. N-free extractives gave a relatively poor estimate of the digestible carbohydrate, whilst both % of sugar + starch and % of available carbohydrate were good measures of digestible carbohydrate.
A. H. CORNFIELD.

**Digestibility of the carbohydrate complex by birds of different ages.** W. Bolton (*J. agric. Sci.*, 1955, **46**, 420—424).—By conventional methods of analysis the apparent digestibility of the crude fibre in compounded poultry diets depended on differences in the composition of the crude fibre in the diet from that in the droppings (the latter contained a larger proportion of cellulose). Cellulose and lignin were not digested. The digestibility of pentosans increased with the age of the birds. Birds of all ages digested sugar and starch. The " available carbohydrate " (sugar + starch + dextrin, expressed as starch) × 1·1 agreed closely with the digestible carbohydrate determined by direct digestibility trials and can be used to calculate the nutritive ratio of a mixed diet and the protein supplement required. A. G. POLLARD.

**Experimental production of " green yolks " by oral administration of sodium copper chlorophyllin.** A. N. Worden, J. Bunyan, A. W. Davies and M. Kleissner (*J. agric. Sci.*, 1955, **46**, 384—385).—The natural occurrence of " green yolks " in eggs was simulated by addition of a water-sol. Na Cu chlorophyllin prep. to the ration of Light Sussex hens. Affected yolks contained ~1 mg. of the chlorophyllin. No such effects were obtained by administration of an oil-sol. prep. of Cu phæophytin. A. G. POLLARD.

**Relationship between riboflavin, hatchability and clubbed down.** R. Coles and F. Cumber (*J. agric. Sci.*, 1955, **46**, 191—198).—The hatching rate of fertile eggs from birds having access to grass was about 80% for rations containing riboflavin (1·6—14·2 μg. per g. of feed). There was only a slight improvement on the highest level of riboflavin. Lack of vitamin $B_{12}$ or an unidentified factor(s) in the APF supplement may be the reason for hatchability not exceeding 80%. The incidence of clubbed down among dead-in-shell decreased with increasing riboflavin content of the dams' diet.
A. H. CORNFIELD.

**Relationships among physical, functional and flavour properties of eggs.** B. A. McLaren and W. J. Stadelman (*Wash. agric. Exp. Sta.*, 1954, *Tech. Bull.* 14, 31 pp.).—The relationships between various methods of evaluating physical quality of eggs and their functional attributes and culinary quality are compared. The effect of breed, type of diet, length and duration of egg storage, and effect of oiling prior to storage on these properties are also reported.
A. H. CORNFIELD.

**Sexual maturity and related phenomena in the domestic fowl.** E. S. E. Hafez and G. A. R. Kamar (*J. agric. Sci.*, 1955, **46**, 9—18).—In Egypt Fayomi pullets hatched from Dec. to Feb. took longer to reach sexual maturity and had a lower body wt. at maturity than had birds hatched from June to Aug. The period over which the first 10 eggs were laid was greater with the summer-hatched birds. Summer and winter hatches attained sexual maturity at an older age and heavier body wt. than did autumn (Sept.—Nov.) hatches. Growth rate of summer hatches was slightly greater than that of winter and autumn hatches whilst that of spring hatches was rather lower. Most of the first eggs laid were non-fertilisable or non-hatchable. The chances of the first egg being fertilisable and hatchable increased with age and wt. of dams and wt. of egg.
A. H. CORNFIELD.

**Time of passage of feed through [alimentary canal of] silver fox.** R. Neseni, M. Lecht and B. Scheven (*Arch. Tierernähr.*, 1955, **5**, 26—32).—For different breeds, times of complete passage are for 1 meal per day 30—32 hr., and for two meals, 10 hr. for the morning, and 29—31 hr. for the afternoon meal. Excretion of ~57% of the meal occurs 10—20 hr. after intake. Approx. 90% of the feed is found in the stomach 8—10 hr. after intake. Rates of passage of the ingesta are 6·5—12 cm. per hr. for solid, and ~90 cm. for fluid components. P. S. ARUP.

**Diseases of animals. I.** A. W. Stableforth. **II. Biochemistry and animal health.** J. L. McGirr (*J. roy. agric. Soc.*, 1954, **115**, 184—192, 192—198).—I. A review of recent work on Johne's disease, bovine mastitis, pneumonia and hyperkeratosis in cattle, pulpy kidney and hæmolytic anæmia in sheep, coccidiosis in poultry and myxomatosis in rabbits.

II. A summary of research progress in hypomagnesæmia, Cu deficiency, acetonæmia, pregnancy toxæmia and fluorosis. Toxic hazards of pesticides are noted. A. G. POLLARD.

**Lead poisoning in cattle.** C. P. Craven (*Qd agric. J.*, 1954, **78**, 41—44).—Possible sources of Pb resulting in poisoning of cattle are discussed and symptoms and treatment are described.
A. H. CORNFIELD.

**Fluorosis of merino sheep in Queensland.** J. M. Harvey and G. R. Moule (*Qd agric. J.*, 1954, **79**, 291—298, 357—359).—The F content of waters from artesian and sub-artesian bores over most of Queensland is shown. Most waters contained < 2 p.p.m. F, whilst some water contained > 10 p.p.m. F. The teeth of sheep drinking water containing < 2 p.p.m. F were normal, with 2 p.p.m. F-containing waters mild toxicity symptoms occurred, whilst the extent of the symptoms increased with the F content of the water drunk by the sheep. Ewes drinking waters containing F pass some of the element on to their lambs. Normal lambs were produced providing their dams consumed waters containing < 10 p.p.m. F. No practical method of treating water on a large scale to eliminate the F hazard is yet available, although small quantities can be treated with $CaSO_4$. F-toxicity symptoms were no less in sheep which received supplements rich in CaO, P and/or protein than in those which received no supplements. A. H. CORNFIELD.

**Fluorosis in cattle in the Northern Province of Tanganyika.** G. W. Walker and A. H. Milne (*E. Afr. agric. J.*, 1955, **21**, 2—5).—The F content of river waters in the area ranged from 1·1 to 45·5 p.p.m. In areas where the values ranged from 14·3 to 45·5 p.p.m. cattle showed dental lesions and other F toxicity symptoms.
A. H. CORNFIELD.

**Selenised wool: preliminary study.** R. O. Leonard and R. H. Burns (*J. Anim. Sci.*, 1955, **14**, 446—457).—The Se contents of the blood and wool of sheep grazing on seleniferous pasture are recorded. No relationship was apparent between wool-Se and blood-Se. Se had no significant effect on the length, thickness or strength of wool fibres but the Se content of the wool was statistically correlated with the no. of distorted fibres. A. G. POLLARD.

**Arsenical poisoning of stock.** R. G. MacDonald (*Qd agric. J.*, 1954, **78**, 114—116).—Sources of arsenical poisoning are discussed. Symptoms, diagnosis and methods of treatment are described.
A. H. CORNFIELD.

**Toxicity of some arsenicals fed to growing-fattening lambs.** L. L. Bucy, U. S. Garrigus, R. M. Forbes, H. W. Norton and W. W. Moore (*J. Anim. Sci.*, 1955, **14**, 435—445).—In self-feeding trials varying proportions of As (0·05—0·4% of the ration) were given in the form of K arsenite, arsanilic acid or 3-nitro-4-hydroxyphenylarsonic acid. The last-named was the most palatable and the least toxic. Pathological effects on liver, kidneys and blood are described.
A. G. POLLARD.

**Coccidiosis in chickens.** J. O. Heishman, C. J. Cunningham and T. B. Clark (*West Virginia agric. Exp. Sta.*, 1955, *Bull.* 376, 11 pp.).—Addition of 0·0062—0·0125% of sulphaquinoxaline or 0·1025% of nitrophenide ("Megasul") to the diet of broilers throughout their life had a beneficial effect on livability, body-wt. gains and feed utilisation. A. H. CORNFIELD.

**Bloat investigations.** W. S. Ferguson and R. A. Terry (*J. agric. Sci.*, 1955, **46**, 257—266).—Bloat was produced in dairy cows and sheep by administration of lucerne juice. Administration of quercetin and/or KCN did not produce bloat in sheep. The bloat factor in lucerne juice was not removed when chloroplastic material was pptd. and when the juice was passed through anion or cation exchange resins. Dosing sheep with lucerne saponins, other saponins, egg albumin or a synthetic foaming compound did not produce bloat. Two household detergents and a no. of surface-active agents did not relieve bloat. A foam-breaking substance (Avlinox, a polyoxyethylene derivative of ricinoleic acid) prevented bloat from developing and also cured a severely-bloated sheep. Administration of synthetic and natural saliva did not affect severity of bloat. A. H. CORNFIELD

**Genetic and alimentary factors and infections.** G. Piana and F. Usuelli (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 287—372).—Reduced calorific value of foodstuffs results in lowered resistance of animals to bacterial infection, but varying response to virus infections. Production of antibodies suffers if the organism cannot draw upon stored intracellular globulins. The results of lipin-deficiencies are often traceable to deficiency of lipo-sol. vitamins. Resistance to infections also results when mineral deficiencies act prejudicially on protein and vitamin action. Shortage of vitamins causes interference with protein metabolism and antibody synthesis, and has more specific effects in the case of vitamins C, A, E, etc. There is hereditary resistance as well as hereditary receptivity, and such resistance may be a feature of breed, strain and family, as well as selection. Evidence for hereditary transmission of active immunity is far from conclusive, and complicated by pseudoheredity, or passage of antibody across the placental barrier. F. R. PAULSEN.

**Insecticides for the control of cattle tick.** Anon. (*Qd agric. J.*, 1954, **78**, 207—211, 285—289).—Laboratory and field trials with dipping vats and spraying fluids showed that DDT (0·5%) and $C_6H_6Cl_6$ (0·05%) had slightly irregular effects in controlling ticks. Both materials have remained toxic in vats for up to five years. Although DDT destroys ticks slowly its residual effect is high; the residual effect of $C_6H_6Cl_6$ is somewhat lower. Ticks may develop resistance to $C_6H_6Cl_6$. In both dipping and spraying trials toxaphene (0·5%) was a little more certain in its effects than was DDT or $C_6H_6Cl_6$ and remained stable for three years. Calves under three months of age have been poisoned by toxaphene in some cases. There is little evidence yet of ticks developing resistance to toxaphene. Both chlordane (0·25%) and Dieldrin (0·05%) have given good control as spraying fluids over three years. Both kill ticks quickly and have a fairly high residual effect. Menthachlor, heptachlor, parathion and Aldrin have given good results in laboratory tests but have not yet been tested in the field. A. H. CORNFIELD.

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**Soil-conditioning agents and methods of preparing the same.** Monsanto Chemical Co. (B.P. 731,052, 14.3.52. U.S., 2.1.52).—A new and improved soil-conditioning composition comprises a finely divided solid copolymer of vinyl acetate with a ($C_{\geq 3}$) alkyl (Me) half-ester of maleic acid and maleic anhydride in the form of a water-sol. partial Ca salt (together with a diluent or extender, e.g., water, peatmoss, limestone, gypsum, mineral fertiliser or silage). The partial Ca salt may be pre-formed or a mixture of the finely divided copolymer and sufficient of a Ca compound may be blended together in forming the composition. Soil may be conditioned, e.g., by incorporating $\geq$2% on the wt. of the soil of the pre-formed Ca partial salt; or by incorporating, simultaneously or successively, in a total amount of $\geq$2% on the wt. of the soil, the finely divided polymer and finely divided CaO, $Ca(OH)_2$ or $CaCO_3$; in either case the amount of actual polymer incorporated is 0·01—0·2% of the wt. of the soil. H. L. WHITEHEAD.

**Improvement of soil structure.** Monsanto Chemical Co., Assees of D. T. Mowry and R. M. Hedrick (B.P. 730,463—4, 16.2.51. U.S. 8.3. and 27.11.50).—Soil structure is improved by incorporation of 0·001—2 wt.-% of (A) a co-polymer, mol. wt. $\leq$10,000 (preferably >15,000) of a substituted vinyl compound (styrene, vinyl chloride or acetate, etc.) with a substituted maleate deriv., e.g., maleinamide, alkali metal maleate, substituted-amino-ethyl maleate; (B) an acrylic polymer, mol. wt. $\leq$10,000, or a co-polymer with a vinyl compound (vinyl acetate or acrylonitrile), e.g., the Na salt of a hydrolysed polyvinyl cyanide. The polymer may be used in admixture with a carrier (peat moss, $CaCO_3$, $SiO_2$ or silage) or a fertiliser. F. R. BASFORD.

**Sintered phosphatic fertiliser.** T. Yamaguchi (B.P. 733,228, 16.4.52. Jap., 23.4.51).—Phosphatic fertiliser containing $Ca_3(PO_4)_2$ in α-form, sol. in citric acid, is made by sintering $CaF_2$-containing phosphate rock with steam and $Na_2SO_4$ (or $Na_2CO_3$ or $Na_3PO_4$, in amount ≡ 1 or 2 mol. proportions to $CaF_2$ present), adding sufficient C-containing material to decompose $Na_2SO_4$ to $Na_2SO_3$ (+ CO), and $SiO_2$, if necessary, to convert CaO formed to $CaSiO_3$. J. S. C.

**Alkylamine salts of α-2 : 4 : 5-trichlorophenoxypropionic acid.** Dow Chemical Co. (Inventor: Bill M. Williams) (B.P. 731,902, 23.10.53).—α-2 : 4 : 6-Trichlorophenoxypropionic acid (**I**) is added to excess of $NR_2R'$ (R is H or as R', R' is alkyl of 1—4 C), to give the amine salt, sol. in water insol. in non-polar solvents and useful as component of plant growth-regulating compositions. Thus, **I** (27) is added to $CO_2$-cooled dimethylamine (10), then excess of amine is distilled off, leaving the *dimethylammonium salt* (31·5 g.), m.p. 113—115°, of **I**. The prep. of the *methylammonium*, m.p. 198—199°, iso*propylammonium*, m.p. 177·5—179·5°, *diiso*propylammonium, *triethylammonium* and sec.-*butylammonium* salts of **I** is also described. F. R. BASFORD.

**Compositions for the treatment of seeds.** Soc. des Usines Chimiques Rhône-Poulenc (B.P. 730,253, 15.5.53. Fr., 12.6.52. Addn. to B.P. 688,736, 27.3.51).—A composition suitable for the treatment of seeds to protect them against crows comprises a di- or tri-aryl (di-*o*-tolyl or di- or tri-phenyl)guanidine (**I**) dissolved in commercial grade oleic acid. The protection may be obtained by coating or impregnating the seed with the compositions containing 10—14% by wt. of **I**, applied to leave 60—210 g. of **I** per 100 kg. of seed. The compositions are not washed out by rain, and they remain effective for long periods without retarding germination of the seed. H. L. WHITEHEAD.

**Tetrahydrofurfuryl ester of 4-chloro-2-methylphenoxyacetic acid as herbicidal agent.** California Spray-Chemical Corp. (B.P. 731,986, 24.2.53. U.S., 14.5.52).—The *ester*, b.p. 179—181°/1 mm., $n_D^{22}$ 1·5271, is characterised by low volatility and is thus more suitable for use as a herbicide than the conventional lower-boiling esters. The ester is made from the appropriate acid and ester in presence of benzene and toluene-*p*-sulphonic acid. A suitable formulation is given. F. R. BASFORD.

**Parasiticidal compositions.** U.S. Rubber Co. (B.P. 731,141, 10,11.53. U.S., 12.11.52).—An effective pesticide composition comprises a chloroalkyl compound (1—150), e.g. a chloroalkyl-aryloxyalkyl sulphite (2-chloroethyl 2-*p*-*tert*.-butylphenoxy*iso*propyl sulphite, Aramite) with Aldrin, BHC, Dieldrin, chlordane, DDT or *N*-trichloromethylthio-tetrahydro-phthalimide (Orthocide), Gammexane, or DDT; powdered mineral silcate (**I**; 100 pt.), and solids (0·5—15 wt.-% on **I**) of the alkaline effluent from the fractional pptn. of waste sulphite liquor (ex the manufacture of wood pulp). F. R. BASFORD.

[**Compounding of**] **weed-killers.** Borax Consolidated, Ltd. (Inventor: Harold P. Knight)( B.P. 730,212, 16.9.52).—A mixture of granular $NaClO_4$, granular hydrated Na borate (borax, optionally partly dehydrated, or partly dehydrated Na metaborate), and optionally $H_3BO_3$, is heated just above the transition temp. of Na borate, with liberation of part of the water of crystallisation, then the mixture is cooled (with absorption of water), and pulverised, to give a granular weed-killer. F. R. BASFORD.

**Retardation of sprouting of seed potatoes.** N.V. Philips' Gloeilampenfabrieken (B.P. 732,163, 20.3.53. Neth., 25.3.52).—The seed is dressed with 1 : 1 : 2 : 3 : 3 : 4 : 5 : 5-octachloropent-1-ene (20—80 mg. per kg.), diluted with inert solid carrier (talc, etc.). F. R. BASFORD.

[**Device**] **for introducing a predetermined amount of a poisonous material [especially fluoro- or azido-phosphine oxides] beneath the surface of the soil.** Pest Control, Ltd. (Inventor: G. S. Hartley) (B.P. 730,587, 16.6.52). F. R. BASFORD.

**Apparatus for distribution of liquids in soils.** Imperial Chemical Industries Ltd. (Inventors: H. R. Jameson and Lenard G. James) (B.P. 729,771, 29.9.52). J. ROBERTS.

**Dry separation of materials, [e.g., tobacco leaf from stalk].** Vokes, Ltd. (Inventor: L. E. R. Umney) (B.P. 730,200, 27.6.52).—The mixture is continuously fed on to a perforated conveyor section where it is subjected to an air stream flowing from a lower to an upper air duct. The lighter component particles are picked up and carried to a centrifugal separator by way of a reduced corner piece and a horizontal section of the duct. The separated particles are continuously extracted through a rotary sealing valve and fed down a shoot on to a conveyor. A draught-inducing fan re-circulates the air stream whilst a small proportion of the latter is passed through a filter to remove dust. J. ROBERTS.

**Production of animal feeding stuffs.** Alginate Industries, Ltd. (Inventors: R. R. Merton and R. W. Moncrieff) (B.P. 730,874, 20.5.52).—Calf meal, to be used in the form of an aq. suspension, is compounded with ⩽1 wt.-% of a mixture of alginic acid (3) and the stoicheiometric amount of a base, e.g., $NaHCO_3$ (1 pt.), to serve as dispersing agent. A small amount of $NaPO_3$ may also be used.
F. R. BASFORD.

**Milk products for animal feeding.** Chas. Pfizer & Co., Inc. (B.P. 730,568, 11.3.52, U.S., 21.4.51).—A stable, synthetic milk product for feeding young animals is prepared by blending dry skim milk (66·6), lard (25·0), fish solubles (12·5), lecithin as emulsifier (4·0 lb.), oxytetracycline as antibiotic (240 g.), vitamin $B_{12}$ (1 lb.), and a number of other ingredients, including vitamins and trace minerals (Co, Fe, Mn, Cu, Zn, Sn), and suspending the mixture in 5 pt. of warm water by weight. L. S.

**Silos.** A.-B. Standardhus (Inventors: S. A. Lundgren and E. B. G. Berg) (B.P. 730,516, 18.6.53).—Since the main cause of pressure on the walls of a deep silo is due to the frictional load, the introduction of members which increase the internal periphery of the silo lessens the pressure and the silo wall is relieved. These elements are vertical plates fixed in various positions in the silo.
K. RIDGWAY.

## 2.—FOODS

**Hygroscopic equilibria of rough rice at elevated temperatures.** J. T. Hogan and M. L. Karon (*J. agric. Food Chem.*, 1955, **3**, 855—860).—Hygroscopic moisture relationships of rice were studied to supplement published data and to furnish hygroscopic equilibria data over a range of relative humidities at 80—115°F. for moisture contents of 11—22% dry basis. These data are examined and correlated. Adsorption of water on rough rice occurs in three stages: a moisture content of 0—7% represents a unilayer of water mol.; the second is characterised by addition of an equal no. of mol. to the already adsorbed unilayer; and the third is a multilayer addition of water from approx. 14% moisture content to saturation. (17 references.)
E. M. J.

**Polysaccharide content of oats, *Avena sativa* L.** D. M. W. Anderson and C. T. Greenwood (*J. Sci. Food Agric.*, 1955, **6**, 587—592).—The kernels of oats were ground, de-fatted with boiling benzene–methanol, and subjected to successive extractions with cold water (five treatments), hot water (90°, five treatments), 5% w/v aq. NaOH at room temp. (five treatments), and aq. NaOH at 90° in a $N_2$ atm. The fractions were analysed for sugars, protein and ash. The starch was purified by repeated dispersal in a mixture of 1M-aq. NaCl and toluene. Oxidation of the unfractionated starch with K metaperiodate showed the ratio of terminal to non-terminal glucose units to be 1 : 27·4. Differential potentiometric I titrations indicated 26·0% of amylose in the starch; the average length of unit-chain in the amylopectin component was hence calculated as 20·3 glucose units. (24 references.) J. S. C.

**Terminal amino-acids of wheat gliadin.** L. K. Ramachandran and W. B. McConnell (*Canad. J. Chem.*, 1955, **33**, 1463—1466).—Wheat gliadin has been found by two different methods to contain three *N*-terminal histidine residues for each mol. wt. of 27,000. Trace amounts of *N*-terminal aspartic acid, glutamic acid, alanine valine and serine were also detected in the preparation used. Hydrolysis in boiling HCl destroyed 5—25% of the bis-(2 : 4-dinitrophenyl) deriv. of histidine, depending upon the time and conditions. Carboxypeptidase did not release free amino-acids from wheat gliadin, but qual. evidence indicates that glutamic acid and leucine occupy the *C*-terminal positions. O. M. WHITTON.

**Starch structure. I. Analytical methods.** C. H. F. Fuller, L. H. Lampitt and L. Coton. **II. Determination of iodine absorption by amperometric titration.** L. Coton, L. H. Lampitt and C. H. F. Fuller (*J. Sci. Food Agric.*, 1955, **6**, 656—660, 660—664).—I. A scheme for the examination of a starch fraction is described. A solution of starch fraction in dilute alkali is prepared and stored at low temp. in a $N_2$ atm.; no measurable change occurs during several days. After determination of polysaccharide concentration, suitable dilutions of the solution are used for several different determinations, e.g., reducing power, limiting viscosity no., properties of the polysaccharide–iodine complex and the limit of conversion by β-amylase. (12 references.)

II. An apparatus for the determination of the I absorption of starches and starch fractions by an amperometric method is described and illustrated. By this method the source of the starches may be identified, and, in starches in the granular state, damaged granules give increased absorption. Wheat flours have an absorption curve similar to that of the extracted starch so that the method may be applied directly to the detection of overmilling in flours. E. M. J.

**Determination of starch and sugars in cereals using the anthrone reagent.** K. M. Clegg (*Biochem. J.*, 1955, **61**, Proc. vii).—The colour reaction of anthrone with carbohydrates is modified to give a simple and rapid method for the determination of starch and sugars in cereals. The sugars are extracted with 80% ethanol and the starch in the residual material is extracted with 52% $HClO_4$. The ethanol is evaporated from the sugar extracts, and aliquots of the dil. starch and sugar extracts are used for analysis. The starch contents of cereals and cereal by-products show a standard deviation of 2·2% of the mean. The results are of use in assessing the metabolisable energy value of feeding stuffs for poultry. Presence of non-cereal material in the sample does not interfere with the analysis.
J. N. ASHLEY.

**Activity of an amylase derived from *Bacillus macerans*.** F. Cramer and D. Steinle (*Liebigs Ann.*, 1955, **595**, 81—100).—The decomposition of native starch by an amylase derived from *B. macerans* is studied, the intermediate and end-products being separated and identified by quant. circular paper chromatography using a 2 : 1 : 1 butanol–dimethylformamide–water system. After separation the paper is treated with aniline phthalate, and the separated constituents localised by examination under u.v. light. The results show that the enzyme acts catalytically and results in the formation of *cyclo*dextrin directly from amylose in a reversible manner. Pure *cyclo*dextrin remains unaffected by the enzyme, unlike glucose, maltose or their homologues which are degraded. A cyclic system is proposed for the complete decomposition of starch by the amylase. (42 references.) G. R. WHALLEY.

**Micro-determination of ester sulphate and free sulphate ions.** P. W. Kent and M. W. Whitehouse (*Analyst*, 1955, **80**, 630—631).—A method is described for the determination of esterified sulphate in sulphated muco-substances, applicable to 2 μg. amounts of $SO_4''$. The mucins or sulphated polysaccharides are heated with formic acid in sealed tubes at 105° for 16 hr. Aliquots of the filtered hydrolysates (or solutions of $SO_4''$ in formic acid) are treated with a benzidine reagent and maintained at 0° for 12 hr. The ppt. is collected by means of immersion filters, washed free from benzidine and dissolved in dil. HCl. The solution is diazotised, coupled with thymol and the colour is measured absorptiometrically, Ilford No. 603 filters (max. transmission at 485 mμ.) being used. The calibration graph is prepared by diazotising and coupling known amounts of benzidine. Pptn. of benzidine sulphate is quant. in the range 2—30 μg. of $SO_4''$. For 5—10 μg. the accuracy is within 10%, and for 10—20 μg. within 5%. When the method was tested with $Na_2{}^{35}SO_4$ and radioactive chondroitin sulphate, all the measurable activity was in the final diazotised solution. A. O. JONES.

**Influence of acids and mineral salts on the Alveographic indices of flours.** —. Margulis and Y. Campagne (*Industr. aliment. agric.*, 1955, **72**, 485—492).—The fixation of ions on gluten influences the tenacity and drawing capacity of the dough. Each anion and each cation carried in electrolytes added to the flour exerts a specific influence. E.g., NaCl has a positive effect on both tenacity and drawing capacity of dough since Cl′ is one of the anions which offers the least opposition to the action of $Na^+$. Gluten behaves as an amphoteric colloid; addition of NaCl results in formation of a Na chloroglutenate of greater elasticity and more capable of stretching than the original gluten. J. S. C.

**Conservation of partially de-oiled groundnut flour.** J. Xabregas, J. Lessian and Y. Bagot (*Oléagineux*, 1955, **10**, 681—685).—Of the various products investigated for preventing oxidation in flour obtained from pressed groundnuts, it was observed that oat flour was only effective with addition of butylated hydroxyanisole. By using natural products, e.g., maize oil or maize germ flour, results comparable with those obtained with similar admixtures of legislatively prohibited synthetic products are obtainable. Maize flour, suitably mixed with pressed-groundnut flour, provides a well-balanced product both as regards amino-acid and vitamin contents, suitable for human nutrition. Maize flour supplements the degerminated flour of groundnut and conserves the oil content. (18 references.) J. S. C.

**Factors that affect the respiration rate of sugar beets.** M. Stout (*Proc. Amer. Soc. Sugar Beet Technol.*, 1954, **8**, 404—409).—The respiration rate of stored sugar beets was reduced by small amounts of $CO_2$ in the atm.; 12% $CO_2$ reduced the $O_2$ uptake by 17%. The $O_2$ concentration and the respiration rate indicated a linear relationship down to about 5% $O_2$; below that concentration some anaërobic respiration took place. Small beets had a greater respiration rate than large beets. Polyploid beets had a lower respiration rate than that of normal diploid varieties. SUG. IND. ABSTR. (E. M. J.).

**Maceration investigations. I. Influence of quantity and temperature.** Anon. (*Tech. Rep., Sugar Res. Inst., Queensland*, 1955, No. 26. From *Int. Sugar J.*, 1955, **57**, 381).—A summary is given of the

maceration tests conducted through the 1954 crushing season at three sugar mills. The optimum maceration was determined as 240% on fibre. There was a positive correlation between maceration quantity and moisture in final bagasse; at one mill this rose from 47·4% at 200% maceration to 50% at 285% maceration. High temp. maceration at one mill gave an increased extraction of 0·4% over warm maceration. Where the temp. of the maceration fluids fell below 160°F. no benefit from heat was recorded. Tests were made at mills without pressure feeders, and with hydraulics on only the final mill of one train. There were higher bagasse moistures at intermediate and final mills. Best results are indicated with mills provided with pressure feeders to reduce bagasse moistures.

SUG. IND. ABSTR. (E. M. J.).

**Effect on juice purity of the removal of ammonia and carbon dioxide in beet sugar factory evaporators.** A. Carruthers and J. F. T. Oldfield (*Int. Sugar J.*, 1955, **57**, 309—310).—Invert sugar concentration of juice was increased by 0·1—0·5 per 100° Brix as a result of evaporation conditions causing decomposition of sucrose; for inversion up to 0·2 per 100° Brix no purity decrease was measured; for an inversion of 0·5 per 100° Brix, the purity drop was only 0·2 units. The addition to thin juice of invert sugar ≡ ~0·2—0·4 g. per 100° Brix gave a reduction in apparent purity equal to that calculated theoretically. A weighed amount of $(NH_4)_2CO_3$ was added to a sample of diluted thick juice which was evaporated under vacuum to 50° Brix, then rediluted to the original Brix. Determination of $NH_3$ and $CO_2$ before and after the evaporation indicated that the $CO_2$ and $NH_3$ contents of the juices were reduced during evaporation and the apparent purity was increased. In normal thin juice samples there were purity increases during evaporation; the increase was greater for juice having a lower pH and greater $CO_2$ content. Although sugar is destroyed during evaporation, the apparent purity of the juice is not reduced to an extent equivalent to the sucrose loss.

SUG. IND. ABSTR. (E. M. J.).

**Effect of ammonia in second saturation. Laboratory carbonatation test.** R. Carolan [*Cómhlucht Siúicre Eirreann Teo* (Irish Sugar Co. Ltd.) *Res. Lab. Rept.* **35**, July, 1955. 4 pp.].—In "micro-factory" trials ~10 ml. of 6% $NH_3$ was added per 2 l. of filtered first carbonatation juice. The increase in N-content was 5·4—7·3 mequiv. per 100° Brix. Progressive pre-liming was effected at 40° and main liming at 85°; the juice was evaporated under vacuum. Results are tabulated. The addition of the $NH_3$ reduced the lime salts content to ~0·1 of the usual value; this improvement in lime salts results from the increased $CO_3''$ content, a disadvantage of the $NH_3$ addition being the decrease in pH in evaporation.

SUG. IND. ABSTR. (E. M. J.).

**Automatic control of carbonatation in sugar factories.** J. Genotelle and R. Michel (*Génie chim.*, 1955, **74**, 80—87).—Continuous carbonatation using automatic control is described. Measurement of electrical conductivity is employed in the first stage of carbonatation (the electrical resistance varies by nearly 10% for a variation in alkalinity of 0·010% of CaO), and measurement of pH in the second stage (the same variation in alkalinity is accompanied by a variation of ~0·5 in pH). Continuous carbonatation increases the over-all efficiency of the factory and gives a juice which is easier to filter, but has a slightly poorer colour. This has no effect on the quality of the final product.

J. M. JACOBS.

**Chromatography of disaccharides on a thermocolumn.** Chen-Chuan Tu and Kyle Ward, jun. (*J. Amer. chem. Soc.*, 1955, **77**, 4938—4939).—The separation on a thermocolumn (a heated charcoal column) of a series of pairs of disaccharides, differing either in the component sugars or in the linkage between sugar units, is described. Five pairs of hexose disaccharides studied were lactose and cellobiose, melibiose and cellobiose, lactose and melibiose, cellobiose and gentiobiose, and maltose and cellobiose. The temp. of the column is regulated to the point at which the separation is considered to be optimum. The disaccharides on the column can be completely removed with 3% aq. ethanol by heating.

O. M. WHITTON.

**Biochemical studies on carbohydrates. CLXIII. A spectrophotometric determination of hexoses in sugar mixtures and polysaccharides.** H. Masamune and K. Ogawa (*Tôhoku J. exp. Med.*, 1954, **60**, 11—21).—The absorption spectra of the reaction products of sugars heated with thionalide (2-mercapto-*N*-2-naphthylacetamide) in pure conc. $H_2SO_4$ for 3 min. were examined. Galactose gives intensive absorption with max. at 605 and 495 mμ.; glucose, fructose and mannose compounds give a max. at 495 mμ. only.

SUG. IND. ABSTR. (E. M. J.).

**Spectroscopic study on the interaction of amino-acids and sugars.** Y. Nagai (*Tôhoku J. exp. Med.*, 1955, **61**, 331—337).—The compounds produced by the reaction of glucose, galactose or arabinose with various amino-acids were made to react with *p*-dimethylaminobenzaldehyde (Ehrlich's reagent) to give dyes, the colour absorption curves of which were examined. The α-amino-acid–sugar compounds gave dyes with max. ~520 mμ. If the α-amino-acids had a second amino-group, the max. was ~576 mμ. These results are compared with absorptions of various other compounds.

SUG. IND. ABSTR. (E. M. J.).

**Effect of cations on reducing-sugar determinations with Shaffer and Hartmann's or Somogyi's reagents.** T. A. Kilroe-Smith and J. F. de Gier (*Analyst*, 1955, **80**, 627—629).—The effects of different ions on the Shaffer and Hartmann micro-reagent and on the Somogyi reagent (compositions described) have been investigated. Three different salts ($CaCl_2$, $MgCl_2$ and $FeCl_3$) were used and all had a profound effect on the reduction of glucose with each reagent. The results indicate that the effects noted are probably due to a change in the buffering power of the oxidising mixture, since the cations are all able to form ppt. with the reagents. The concn. of these cations should therefore be kept at a min. in reducing-sugar determinations with these reagents. $Fe^{***}$ is particularly troublesome, probably as the result of its oxidising power. Similar effects probably occur with any cations which alter the buffering capacity of the reagent. The effect of varying concn. of $MgCl_2$ on the activity of cellulase from a strain of *Hydnum henningsii* is shown.

A. O. JONES.

**Unfermentable sugars. IV. Action of β-glucosidase on the unfermentable disaccharides. V. Isolation of *iso*maltose and saké-biose from koji extract.** K. Matsuda and K. Aso. **VI. Oligosaccharides synthesised from maltose by *Schizosaccharomyces pombe*. VII. Synthesis of *iso*maltose from glucose.** K. Shibasaki and K. Aso (*Tohoku J. agric. Res.*, 1954, **5**, 123—125, 125—129, 131—138, 138—142).—IV. Two sugars previously reported (Aso and Shibasaki, *ibid.*, **3**, 237) as present in koji extract after fermentation are shown to be unattacked by β-glucosidase (emulsion). They are designated saké-biose (probably 3-*O*-α-D-glucopyranosyl-D-glucopyranose) and koji-biose.

V. Residual sugars present in fermented koji extract, separated by chromatography and isolated as cryst. octa-acetates, included *iso*-maltose, maltose and saké-biose.

VI. *Schizosaccharomyces pombe* was suspended in buffered (pH 4·7) maltose solution and incubated 1—2 hr. at 28—30°. Saké-biose, koji-biose, *iso*maltose, panose and dextrantriose were produced (quant. data recorded). *S. pombe* also produced *iso*maltose from glucose or sucrose.

VII. Culture filtrates from *A. niger* added to buffered aq. glucose produced (after 72 hr. at 55°) *iso*maltose, saké-biose and koji-biose.

A. G. POLLARD.

**Unfermentable sugars. VIII. Identification of saké-biose, nigerose and γ-sugar.** K. Matsuda, G. Hiroshima, K. Shibasaki and K. Aso (*Tohoku J. agric. Res.*, 1954, **5**, 239—242).—Nigerose (from *Aspergillus niger*), saké-biose (Matsuda and Aso, *ibid.*, **5**, 123) and γ acetate (A. Thompson *et al.*, *J. Amer. chem. Soc.*, 1954, **76**, 1309) are identical, viz., 3-*O*-α-D-glucopyranosyl-D-glucopyranose.

A. G. POLLARD.

**Change in status of glucose [as food ingredient]?** C. Nieman (*Conserva*, 1955, **4**, 114; cf. *ibid.*, 42).—Recent Dutch legislation and current opinion lay stress on the quality of fruit used in jam making without restrictions on the use of glucose syrup. In any case, glucose is formed from the sucrose during jam boiling.

P. S. ARUP.

**Nutrient content of cane and beet sugar products.** W. A. Krehl and G. R. Cowgill (*Food Res.*, 1955, **20**, 449—468).—Representative samples (~240) of various sugar products were collected from various localities and assayed for: thiamine, riboflavin, pyridoxine, niacin, pantothenic acid, biotin, folic acid and inositol. These and other data on inorg. constituents and amino-acids are presented. The vitamins present are in quantities so small as to be completely impracticable as a significant nutritional source of these substances. (49 references.)

E. M. J.

**Examination of Scottish heather honey. II.** T. J. Mitchell, L. Irvine and R. H. Scoular (*Analyst*, 1955, **80**, 620—622).—Previous examination of 42 samples of honey, of which 30 were predominantly ling honey, showed evidence of a relationship between the colloid content, total N and thixotropy (Mitchell *et al.*, J.S.F.A. Abstr., 1954, ii, 236). Since the high colloid content of ling honey has been shown to be the cause of thixotropy (Pryce-Jones, *Proc. Linn. Soc. Lond.*, 1944, **2**, 129) the nature of the colloid is of interest. In 25 samples of ling honey and one of bell-heather honey, the N content, pH, moisture and thixotropy were determined. The pH ranged from 4·20 to 5·36 for the ling honeys, whereas the bell-heather honey had a pH of 4·14. The thixotropic ratio ranged from 1 to 100, the bell-heather honey having poor thixotropy. On a dry basis the colloid pptd. by trichloroacetic acid ranged from 1·35 to 3·90% for the ling honeys and was 0·80 for the bell-heather honey. The average amount of protein in the honey colloid was 64%. Although a rough parallelism appears to exist between the thixotropy and the colloid content, no exact relation was found.

A. O. JONES.

**Rapid sugar extraction procedure for analysis of candied fruits, jams and fresh fruits.** J. A. Kitson, C. C. Strachan and R. F. Cain (*J. agric. Food Chem.*, 1955, **3**, 862—864).—A Waring Blendor was used to obtain an aq. solution of the sugars and the technique of Reifer and Melville (*Proc. 11th Int. Congr. Pure and Appl. Chem., Lond.*, 1947, **3**, 223) with two modifications (the non-use of ether during blending, and the filtration of the extract mixed with asbestos through a sintered-glass filter) was employed. The reducing sugars were determined by the Lane and Eynon method and the sucrose after inversion was determined as invert sugar in the total sugar estimation. The time taken is <1 hr. compared with 2·5 hr. taken by the AOAC method. Results of the two methods (which are in good agreement) on nine fruits and two marmalades, are compared. (19 references.) E. M. J.

**Effect of sucrose-invert and high conversion glucose syrups in the preparation of candied cherries.** C. C. Strachan and F. E. Atkinson (*Food Technol.*, 1955, **9**, 518—520).—The effect of sucrose-invert sugars alone and in combination with high conversion type glucose in the prep. of candied cherries by the hot continuous syruping method was studied. A properly balanced sucrose-invert blend (2 : 1 to 1 : 1) resulted in a product of better flavour and less susceptibility to microbial spoilage, and a gain of 10% or more in wt., in comparison with 50% glucose samples, which were of poor appearance, flavour and texture, and no gain in wt., 20% glucose being the max. concentration permitting a satisfactory product. E. M. J.

**Sorbic acid as a fungistatic agent at different pH levels for moulds isolated from strawberries and tomatoes.** E. S. Beneke and F. W. Fabian (*Food Technol.*, 1955, **9**, 486—488).—Data on the effectiveness of sorbic acid in controlling growth of fungi isolated from tomato and strawberry fruits are presented. In tests with varying pH and concentrations of sorbic acid there was slight growth of some strains of *Penicillium, Alternaria* and *Aspergillus* at pH 4·0 and 0·050% of sorbic acid in strawberry puree. Sorbic acid (0·075%) inhibited the growth of all test fungi when added to strawberry puree or tomato juice at a pH natural to these products ; 0·025% inhibited the growth of *Botrytis* and *Rhizopus* in strawberry puree ; and *Colletotrichum, Fusarium, Rhizopus* and *Rhizoctonia* in tomato juice. E. M. J.

**Water-soluble constituents of fruits. II. Separation of acids on anion-exchange resins : isolation of L-quinic acid from apricots.** E. F. L. J. Anet and T. M. Reynolds. **III. Examination of the sugars and polyols of apricots, peaches, pears and apples by paper chromatography.** A. S. F. Ash and T. M. Reynolds. **IV. Water-soluble constituents of fruit.** E. F. L. J. Anet and T. M. Reynolds (*Aust. J. Chem.*, 1955, **8**, 267—275, 276—279, 280—284).—II. Displacement chromatography on columns of strongly-basic anion-exchange resins (Dowex 1-X4, Dowex 2-X4 and Amberlite IRA-400) is used to separate, isolate and purify some water-sol. org. acids. The order of emergence of 27 acids from these columns is recorded. The acids are detected on paper chromatograms by treating with $AgNO_3$ (3 ml. of 50% aq. solution) in acetone (200 ml.) followed by (i) heating the paper for a few min. at 100°, and/or (ii) spraying with ethanol NaOH. The $R_F$ values for 30 acids in two solvent systems are recorded. The method is used for the isolation of L-quinic, succinic, L-malic and citric acids from the apricot-fruit. (23 references.)

III. Paper chromatography is used to illustrate the presence of xylose, fructose, glucose, sucrose, a cyclitol (probably *meso*inositol) and one or more ketose oligosaccharides in apricots, peaches, pears and apples ; and of galactose in pears, and possibly also in peaches and apples. (12 references.)

IV. Displacement chromatography on strongly-basic anion-exchange resins is used to isolate the constituent acids of several varieties of peaches. L-Quinic, L-malic and citric acids are the major constituents, the predominance of any one depending on the variety, season and maturity of the fruit. Mucic acid is found in small quantities and galacturonic acid is only present in fruit picked at commercial maturity and ripened at 20°. The effect of maturity on the three major acids is illustrated ; the immature fruit contains only traces of citric acid. (13 references.) D. Bailey.

**Water-soluble constituents of fruit. V. Sugars and polyols of the apricot fruit.** A. S. F. Ash and T. M. Reynolds (*Aust. J. Chem.*, 1955, **8**, 444—450).—The sugars and polyols of the apricot fruit were separated by chromatography on columns of charcoal and cellulose, followed, where necessary, by paper chromatography. Glucose and sorbitol were separated by chemical methods. Xylose was characterised as the dibenzylidene dimethylacetal, fructose as the 2 : 5-dichlorophenylhydrazone, glucose as the diethylmercaptal, sucrose as the octa-acetate, and sorbitol and *meso*inositol as the hexa-acetates. A no. of apricot oligosaccharides composed of glucose and fructose units were separated by chromatography on charcoal and paper. (27 references.) O. M. Whitton.

**Cultivation of *Torula utilis* in waste lyes from fermentation of citric acid. II. Cultivation on a semi-technical scale.** H. Leopold and Z. Fencl (*Chem. Tech., Berlin*, 1955, **7**, 608—615).—The waste (hitherto useless) lyes resulting from the manufacture of citric acid by fermentation of molasses with *Aspergillus niger* followed by pptn. of the citric acid with milk of lime and separation of the Ca citrate are used for the culture of *Torula utilis*. Descriptions are given of batch and continuous processes for the manufacture of yeast by this culture, and trials of the processes on a semi-technical scale. To avoid infection, acid or alkaline lyes are used and are boiled after addition of superphosphate and nutrient salts. On this scale, however, the infection is a major problem, and occurs strongly even with such precautions and when keeping the (transport) solutions cold. Vessels of Fe or Al are used, but Fe vessels with acid liquors introduce an Fe content which detracts from the quality of the yeast produced. While the yields of yeast obtained are not as good as those obtained in the laboratory (being reduced largely by the infection) they surpass those obtainable by the use of molasses or sulphite liquor as substrate. The spent liquor remaining, after filtration, from the yeast product is of greatly reduced content of ($KMnO_4$-) oxidisable substances and can readily be purified by chemical or biological means. H. L. Whitehead.

**"Purified" lemon-juice concentrates.** E. Benk (*Riechstoffe u. Aromen*, 1955, **5**, 318—320).—A discussion is given of the constitution of commercial conc. lemon juices current in non-German countries. These juices correspond, in citric acid content, with a concn. ~4—5 times that of the original fruit-juice, but lack the flavour and proper quantity of essential constituents of the fruit-juice (notably sugars, amino-acids, pectins, albumins and minerals). Analyses of typical products are given. The deficiencies of taste and of essential natural constituents are ascribed to a process of manufacture involving treatment with alcohol and evaporation under vac. wherein the alcohol serves to precipitate the essential constituents. A discussion is given of German law covering the manufacture, sale and trade description of the juices. H. L. Whitehead.

**Stabilised lemonade powder.** G. K. Notter, D. H. Taylor and L. H. Walker (*Food Technol.*, 1955, **9**, 503—505).—A method of preparation of a powder, containing only the components of fresh lemonade (lemon solids, lemon oil and sucrose), soluble in cold water, the solution having an appearance of home-made lemonade, is reported. No loss of ascorbic acid occurred during prep. No changes in colour or flavour were found after storage for three months at 70°F. (20 references.) E. M. J.

**Chromatographic comparison of non-volatile acids of fresh and stored apple juice concentrate.** M. L. Buch, E. C. Dryden and C. H. Hills (*J. agric. Food Chem.*, 1955, **3**, 960—964).—No difference was found in the acids of fresh and of storage-darkened apple juice concentrate on separation by chromatographic method. By reference to a table of chromatographic constants of standard acids and apple concentrate acids tentative identification of the following was made : galacturonic, quinic, phosphoric, citric, malic, chlorogenic, citramalic, caffeic, succinic and lactic acids. (19 references.) E. M. J.

**Storage behaviour of powdered apple and grape juice products.** V. A. Turkot, H. I. Sinnamon, R. K. Eskew and G. W. Macpherson Phillips (*Food Technol.*, 1955, **9**, 506—509).—Both powders can be stored satisfactorily for one year at 73°F., and for six months at 100°F. if packed in 4-oz. cans containing a desiccant envelope. The moisture content of the powders packed at 73°F. may be as high as 2·9 for apple, 2·5% for grape and at 100°F., 2·0 for apple and 1·7% or less for grape. There were no significant differences in storage stability when the products were packed in air, $N_2$ or vacuum. E. M. J.

**Effect of processing methods on the colour of tomato juice.** R. B. Davis and W. A. Gould (*Food Technol.*, 1955, **9**, 540—547).—The colour of canned tomato juice is studied in relation to four methods of processing (*a*) "hot-break," high-temp., short-time- ; (*b*) "cold-break," high-temp., short-time- ; (*c*) "hot-break", conventional retort- and (*d*) "cold-break," conventional retort process. The results obtained emphasise the need for taking into consideration the three attributes of colour—hue, value and chroma—and indicate the desirability of further study of the effects of these attributes on visual colour evaluation of tomato products. (19 references.) E. M. J.

**A proposed method for converting Hunter colour difference meter readings to Munsell hue, value and chroma renotations corrected for Munsell value.** R. B. Davis and W. A. Gould (*Food Technol.*, 1955, **9**, 536—540).—The method described is proposed for converting Hunter colour difference meter readings of tomato juice samples to Munsell terms, and increasing the accuracy of interpretations of colour data obtained. (25 references.) E. M. J.

**Phenolic compounds in potato tissue.** R. C. Cheng and F. Hanning (*Food Res.*, 1955, **20**, 506—511).—Tannins (tannin and tyrosine)

were more concentrated in the skin than in the centre flesh of the potato, and more in the stem end than the bud end. By paper chromatographic technique, chlorogenic acid was found in the skin, discoloured areas and the centre flesh; caffeic acid was present in the skin; L-tyrosine was found in the discoloured areas and in the centre flesh. These substances were confirmed by spectrophotometric method. E. M. J.

**Cooking of potatoes for sodium-restricted diet.** A. Schoustra and J. J. L. Willems (*Voeding*, 1955, **16**, 706—708).—The Na content of potatoes is not materially altered after cooking in the unpeeled state together with peeled potatoes and salt. P. S. ARUP.

**Effect of moisture and high temperature on cell walls in plant tissues.** C. Sterling (*Food Res.*, 1955, **20**, 474—479).—Food substances, carrot, potato and apple were subjected to steaming procedures for periods up to an hour and examined microscopically. In no case were cell walls ruptured. Tissue structure was modified only by the separation of intact cells. (23 references.) E. M. J.

**Influence of some metallic ions on vitamins in food. I. Decomposition of carotene in the carrot.** T. Goto and M. Arii (*Tohoku J. agric. Res.*, 1954, **5**, 63—70).—Significant losses of carotene resulted from boiling (20 min.) chopped carrot with Fe, Pb, Mn, K or Na salts (1—10 mg. of base per 100 c.c. of water). Zn and Co salts had no effect. In some cases (Mn) carotene was decomposed by soaking in the cold solution. A. G. POLLARD.

**Adsorptive and ion-exchange power of pectocellulosic vegetable tissues.** R. Cultrera, V. Averna and E. Trifirò (*Ann. Chim., Roma*, 1955, **45**, 854—868).—The adsorption of all the cations examined ($H^{\cdot}$, $Li^{\cdot}$, $Na^{\cdot}$, $K^{\cdot}$, $NH_4^{\cdot}$, $Mg^{\cdot\cdot}$, $Ca^{\cdot\cdot}$, $Ba^{\cdot\cdot}$, $Cu^{\cdot\cdot}$ and $Al^{\cdot\cdot\cdot}$) on pectocellulosic vegetable tissue and exchange between the adsorbed ions and aq. solutions of electrolytes has been demonstrated. Only part of the adsorbed $Cu^{\cdot\cdot}$ was displaced with electrolyte solutions, suggesting two mechanisms of fixation. The adsorptive power of the tissue was not destroyed by dehydration with EtOH or by heat. In the light of the results the mechanism of transport of ions in tissues is discussed. L. A. O'NEILL.

**Effect of degree of enzyme inactivation and storage temperature on quality retention in frozen peas.** W. C. Dietrich, F. E. Lindquist, G. S. Bohart, H. J. Morris and Marvel-Dare Nutting (*Food Res.*, 1955, **20**, 480—491).—Data on peroxidase and catalase assays are presented. There was greater loss of ascorbic acid and degradation of chlorophyll in materials containing active catalase than in those in which catalase and peroxidase had been inactivated. Samples of peas blanched just adequately to the semiquantitative peroxidase test were better in flavour, colour, and contained a greater amount of ascorbic acid than did samples blanched for longer or shorter times. These peas retained excellent quality for a year. Deterioration increased with increasing storage temp. (28 references.) E. M. J.

**Influence of maturity and storage treatments on the ascorbic acid content of the seeds of southern peas.** M. W. Hoover (*Food Res.*, 1955, **20**, 469—473).—Southern peas (*Vigna sinensis*) are a good source of ascorbic acid if used at an early stage in their development, or if they are held in storage at 40°F. If the storage temp. is increased to 70° or 100°F. there is a decrease of 42 and 54% respectively. Prolonging the time of storage had the same general effect as increasing the storage temp. E. M. J.

**Determination of lindane in mushrooms.** I. Hornstein (*J. agric. Food Chem.*, 1955, **3**, 848—849).—The mushrooms (100 g.) are washed, cut into small pieces and refluxed for 3—4 hr. with methylene chloride. After standing overnight the extract is decanted through a filter paper into a separating funnel, the small amount of water is separated and the methylene chloride is reduced in vol. on a water bath to about 50 ml. The extract is then sulphonated using three separate 10-ml. portions of 30% fuming $H_2SO_4$; 25 ml. of cold water are then cautiously added to the methylene chloride, this washing process being repeated with two further 25-ml. portions of water. The methylene chloride is then evaporated to a vol. of 1—2 ml., 120 ml. of glacial acetic acid are added and 20 ml. of the acid are distilled by heating to 130—140° in an oil bath and the glacial acetic solution is analysed. E. M. J.

**Variety, type, year and location effects on the chemical composition of groundnuts.** J. F. Eheart, R. W. Young and A. H. Allison (*Food Res.*, 1955, **20**, 497—505).—Results of analyses of kernels and hay extending over a period of several years, made on some 30 samples of each of a no. of standard varieties of groundnuts, were compared with presently reported composition data on groundnuts. Variety had a greater influence on all the constituents than had year. Year had an important effect on thiamine and riboflavin: location had a greater effect on thiamine, riboflavin and hay protein than had variety: Spanish type groundnuts were higher in protein while the Virginia type was higher in niacin content of kernels and protein in the hay. The variety × year interaction was significant for all constituents except thiamine: mean protein, riboflavin and niacin contents of the kernels were considerably higher, and the mean thiamine content 50% higher than F.A.O. values: protein content of the hay was practically the same as Morrison's value. Variety J-11-L and Introduction 149-637 were highest in nutritive value and could be used in breeding tests to increase nutritive value of high yielding varieties. (16 references.) E. M. J.

**Seaweed as a source of yeast food. I. II.** E. O. Morris (*J. Sci. Food Agric.*, 1955, **6**, 611—618, 618—621).—I. Various extracts and products derived from seaweeds were examined for suitability as media, or media supplements, for the growth of a wide range of yeast genera (46 strains). Effects of environmental variations on the cultures were also studied. Extracts from *Laminaria cloustoni* (frond) provided the most suitable media; the most prolific yeasts were *Candida krusei, Candida solani, Nadsonia fulvescens, Pichia membranæfaciens* and *Oospora lactis*. Aëration of cultures tended to improve yields.

II. The effects of various enzymes in promoting hydrolysis of seaweed polysaccharide fractions were studied. Malt extracts and filtrates of *Myrothecium verrucaria* gave particularly effective results. Seaweeds treated with these enzymes are excellent media for yeast nutrition and fermentation but relatively high enzyme concn. are needed for efficient hydrolysis. J. S. C.

**Gas-tightness of crown cork [bottle caps].** M. Laupheimer (*Brauwelt*, 1955, **95, B**, 1057—1062).—Decreases in pressure in crown-corked bottles containing mineral water are smaller during transport than during storage for an equal period, and are in either case appreciably smaller for bottles kept horizontally than for those kept vertically. Reductions in the moisture content of the cork layer of the caps decrease their gas-tightness. P. S. ARUP.

**Plant engineering memos [Grape juice separation].** E. Mirassou and N. Mirassou (*Industr. Engng Chem.*, 1955, **47**, No. 10, 103A—104A).—Photographs illustrate equipment for automatically separating juice from crushed grapes. Crushed grapes with unfermented juice are carried on a cleat-type conveyor over a screen. The juice drains through and is transferred to the fermenting tanks. The pulp is finally passed to a single screw press. Advantages of the method and of using stainless steel screens are given. O. M. WHITTON.

**Utilisation of the grape. I. Composition of grape juices.** K. Aso, K. Shibasaki, K. Matsuda, T. Nakayama, F. Yamanchi, A. Sasaki, S. Iwasa and S. Hamada. **II. Production of grape juice for drinking.** K. Aso, T. Nakayama, A. Sato and S. Iwasa (*Tohoku J. agric. Res.*, 1954, **5**, 99—105, 107—113).—I. Analyses of commercial and pine grape juices are recorded. Paper chromatography revealed the presence of about 20 amino-acids; of these alanine and glutamine occurred in considerable proportions.

II. The technique of concentrating grape juice is examined in laboratory experiments, attention being given to retention of colour and aroma, clarification and removal of sediments. A. G. POLLARD.

**Utilisation of the grape. III. Inversion of sucrose by grape juice.** K. Aso, T. Nakayama, A. Sato and S. Iwasa (*Tohoku J. agric. Res.*, 1954, **5**, 115—121).—Sucrose added to the juice of some very sour varieties of grapes becomes almost completely inverted. The juice contains a natural invertase which is destroyed at 75—85°. Much of the inversion however, is attributable to org. acids, notably tartaric and malic, the former being the more active. Grape juice is much more active than apple juice in inverting sucrose. A. G. POLLARD.

**Chemical investigation of wine in the pharmaceutical laboratory.** K. G. Bergner and H. Meyer (*Dtsch. ApothZtg.*, 1955, **95**, 977—981, 1009—1012).—A discussion is given of German law governing the manufacture, blending and storage of wines. A description is given of the analytical processes (including rapid methods) used in the pharmaceutical laboratory for testing the wines and the raw and intermediate products (e.g., for alcohol content, sugar content, acidity, fining agents, adulterants and purity) and for detecting contraventions of the Wine Acts. L. H. WHITEHEAD.

**Standardisation of analytical methods for evaluation of activated carbon used in wine and oil industries.** A. Zucca (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 225—233).—Suggested methods for evaluation of activated C for use in wine and oil industries include those for impurities and pH, apparent *d*, effect of carbon on colour, acetic-, tartaric- and volatile-acidity, ash, alcohol, and alkalinity of wines, oleic acid content, peroxide index and Kreis reaction of oils, and ignition loss, ash, pH, matter sol. in water and acid, Fe, Cl′, $SO_4''$, Zn, $PO_4'''$, Ca, Mg and $SiO_2$ of the carbon. F. R. PAULSEN.

**Removal of metals from wines by various methods.** P. G. Garoglio (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52**, Ser. ii, 203—224).—Chemical, physico-chemical and enzymic methods of removing metal ions from wine musts are outlined, with special reference to Fe and Ca. F. R. PAULSEN.

**Pectolytic enzymes for removal of iron in clarification of wine musts.** O. Colagrande (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **51**, Ser. i, 470—483). F. R. PAULSEN.

**Lead content of some Australian wines.** B. C. Rankine (*J. Sci. Food Agric.*, 1955, **6**, 576—581).—The Pb content of 55 Australian wines (including white and red, sweet and dry, fortified and unfortified types) varied from 0·04 to 0·86 (mean 0·23) p.p.m. Results by other workers are reviewed. The present (British) Ministry of Food limit of 1 p.p.m. is discussed and it is suggested that the max. limit should not be reduced below 0·5 p.p.m. J. S. C.

**Evaluation of thresholds and minimum difference concentrations for various constituents of wines. IV. Detectable differences in wine.** E. Hinreiner, F. Filipello, H. W. Berg and A. D. Webb (*Food Technol.*, 1955, **9**, 489—490; cf. J.S.F.A. Abstr., 1955, ii, 241).—Data are presented on the min. detectable concentration differences determined for sucrose, ethyl alcohol, $SO_2$, glycerol, tannin, ethyl acetate, acetaldehyde and tartaric acid in a white and a red table wine. The min. concentration differences for these substances in wine were higher than those in aq. solution. E. M. J.

**Determination of higher alcohols.** J. Mejane and —. Cathy (*Industr. aliment. agric.*, 1955, **72**, 475—480).—Five methods for the determination or detection of higher alcohols in distilled spirits are described and compared. The "official" method consists of a colorimetric test using aniline and $H_3PO_4$, refluxing, and adding conc. $H_2SO_4$. A modification of this uses prepared standards of *iso*butyl alcohol for comparison. The Komarowsky method uses 1% salicylaldehyde and $H_2SO_4$, with comparison tests. It is modified by application to undiluted (96°) spirits (the original method using 50° spirits.) It is further modified by von Fellenberg by replacing salicylaldehyde with *p*-hydroxybenzaldehyde. The results of a series of analyses using these methods are reported; it is concluded that the unmodified Komarowsky method is to be preferred. J. S. C.

**Fine corn distillate in various spirit products.** G. Haeseler (*Riechstoffe u. Aromen*, 1955, **5**, 320—321).—Comparative additions of fine corn distillate (**I**) and of extra fine neutral spirit (from potato) are made to a range of spirit liquors (whisky, brandy, rum, fruit liquors) to test their respective effects on the aroma, taste, etc. In general, the greater enhancing of aroma or improvement is effected by **I**. H. L. WHITEHEAD.

**Tables for determining proteins on dry material in barley and malt, using 1·4 gramme of the sample.** H. Kieninger (*Brauwelt*, 1955, **95, A**, 1017—1019).—The tables are provided as an adjunct to Kjeldahl determinations made on 1·4 g. of the sample with the use of 0·1N NaOH for the titration, and of 30 ml. of 0·1N-$H_2SO_4$ in the receiver for the distillation. Contents of N and proteins as % on the dry material are given for appropriate ranges of burette readings obtained for malt and barley with moisture contents ranging from 3·8—4·3 and 15·0—15·9, respectively. P. S. ARUP.

**Analytical determination of degree of modification of malt.** V. Salač and I. Hlaváček (*Brauwelt*, 1955, **95, B**, 1069—1075, 1098—1100).—Single determinations based on extract differences as between 90 and 40% or 90 and 25% fine meal are not sufficiently accurate for use in practice. The utility of the Kolbach value is limited by several factors. Simplified determinations of Hartong values based on measurements of $\eta$ at 85° (Höppler apparatus) afford the best and most expeditious means for evaluating the degree of modification and liquefying capacity of malts and for checking the progress of malting. A nomogram is given for converting the observed value for $\eta$ to the basis of 10% extract. P. S. ARUP.

**The Bishop value as substitute for barley or malt extract [yields].** F. Kutter (*Schweiz. Brauerei Rdsch.*, 1955, **66**, 159—161).—Data for Swiss-grown barleys show poor agreement between the expected extract calculated according to the Bishop formula, and the corresponding extract % found in laboratory estimations. The Bishop value should be regarded solely as an expression embodying certain properties of the barley, which may, however, be found to have a practical significance when tested against the results of experimental maltings. P. S. ARUP.

**Chemical processes during kilning.** M. Lindemann (*Brauer u. Mälzer*, 1955, **8**, No. 19, 3—8; in English pp. 5—8).—A review covering the regulation of kilning conditions for the production of pale or dark malts and the chemical changes resulting from different treatments. P. S. ARUP.

**Estimation of degree of kilning of light malt.** A. Kaiser and G. Held (*Brauwelt*, 1955, **95, B**, 1211—1215).—Five methods proposed for this purpose are unsatisfactory, but the Hartong and Kretschmer method, based on the ratio between the extracts obtained at 65° and at 80° affords a good criterion. A new method is proposed which is based on the determination of coagulable N in laboratory wort according to Kolbach and Schild, the result being calculated as a % on the Lundin fraction A (instead of % on the extract). The method is shown to give satisfactory results for 16 different malts, and is preferable for convenience. (14 references.) P. S. ARUP.

**Practical experience of the clarification of wort and beer by centrifuging. I. Wort.** N. L. Vacano (*Brass. Malt. belge*, 1955, **5**, 358—368).—The basic principles of centrifugal clarification and the characteristics of separators actually in use for clarification of wort or beer are reviewed. The importance of controlling the rate of sedimentation is shown. The pptn. of particular solid particles is a function of the square of their diameters, and takes place more readily if they are spherical. The sizes of the various particles of substances to be eliminated in brewing are listed; e.g., hot or cold "trub," yeasts, etc. The sedimentation rate also depends on the relative sp. gr. of solid matter and liquor, the viscosity and consistency of the liquor and of its movement. The sp. gr. and viscosity of wort or beer in various stages of the brewing process are given and the sedimentation rates under various conditions are discussed. The reasons for adopting a centrifugal clarification applied immediately after pumping of the wort, to eliminate only the hot "trub," are explained. The efficiency of centrifugal clarification and the extent to which it enables full utilisation of materials to be achieved, both increase with the volume of production, particularly if a continuous cycle of wort clarification can be established. A résumé of actual experience of hot wort clarification and its effects on subsequent stages of brewing and the quality of finished products shows that the beer is purer, paler and in better "biological condition," and that the use of fermenting vats is eliminated with increased wort recovery between brewing house and fermenting cellar. J. S. C.

**Lactic-acid bacteria associated with brewery products. IV. Interrelationships in requirements for purines, folic acid and *p*-aminobenzoic acid.** C. Russell, R. R. Bhandari and T. K. Walker (*J. Sci. Food Agric.*, 1955, **6**, 633—636).—Thirty-four strains of *Lactobacilli*, isolated from beer or growing yeast, were examined in respect of their growth requirements for various substances (cf. J.S.F.A. Abstr., 1955, i, 368); 17 required uracil, none required xanthine. In two cases, growth failed in the absence of guanine and xanthine, suggesting interconversion of these two substances. Requirements varied in respect of both *p*-aminobenzoic acid and folic acid. The results are discussed in relation to those previously obtained. J. S. C.

**Problem of detection of wild yeasts by tartaric acid-wort method.** Ursula Hoffmann (*Brauwelt*, 1955, **95, B**, 1397—1399).—The outcome of the test depends largely on the concn. of the inoculation and the age of the culture. Many wild as well as culture yeasts are more or less inhibited by 1—2% of tartaric acid in the wort, and some undergo transformation into abnormal types. P. S. ARUP.

**Method of operation of the alcohol-dehydrogenases from yeast.** K. Wallenfels and H. Sund (*Angew. Chem.*, 1955, **67**, 517).—The activity of the dehydrogenases (**I**) depends on their content of free SH-groups, increasing to a max. with 36 such groups. With **I** originating from yeast, their operation in the reduction of alcohol to diphospho-pyridine nucleotide (**II**) follows a Meerwein–Ponndorf pattern, but with Zn taking the place of Al in the H transfer and in the formation of the protein–metal–alcohol–**II** complex with hydride H. The greater the no. of free SH-groups the greater is the binding of Zn and likewise the enzymic activity of the compound formed in the substrate. If the SH-value falls below 4, the power of combining with **II** and alcohol, and hence the enzymic power, ceases. H. L. WHITEHEAD.

**The constitution of ribonucleic acid from yeast.** E. Dimroth, H. Witzel, G. Neubauer and D. Matheka (*Angew. Chem.*, 1955, **67**, 518).—The constitution of yeast ribonucleic acid (**I**) is examined from a study of its hydrolysis with aq. $Bi(OH)_3$. Under mild neutral conditions, cyclic $H_3PO_4$ esters are primarily formed by bridging of the ester groups across the 2′- and 3′-C-atoms and simultaneous splitting off of the alcoholic group from the 3′-position, further hydrolysis converting the cyclic ester to a mixture of the isomeric 2′- and 3′-phosphate esters. Under stronger acid conditions the hydrolysis proceeds by saponification of the primary phosphate esters into isomeric di-nucleoside phosphates. A detailed examination of the constitution of the hydrolysis products is made, and the results are examined in the light of the products of hydrolysis by means of ribonuclease. Hence a concept is obtained of the possible structure of **I** and of the mechanism of the hydrolyses. It is thought

that, in the polynucleotide chain, coupling in the normal 3′- and 5′-positions takes place with uridine- (not purine-) nucleotide and with cytidine- (not guanosine-) nucleotide. The occurrence of pyrimidines in the structure is discussed, including the nature and extent of their occurrence in branch chains. No pyrimidines (excepting the grouping cytosine–$PO_4$–adenine) can be found in a purine chain, but 60% of the pyrimidines must occur on the main chain between the point of inversion and the commencement of the purine chain. H. L. WHITEHEAD.

**The pediococcus problem.** E. Schubert and U. Gerhardt (*Brauwissenschaft*, 1955, **8**, 228—235).—Published data on the culture of pediococci are reviewed. The nutrient media at present in use are considered unsatisfactory. An improved medium suitable for the isolation and culture of pediococci consists of broth-agar containing peptone (1%) and NaCl (0·5%), to which 10% of blood is added at 40—50° immediately before use. A simpler medium (with or without agar) suitable for culture, but less so for isolation, can be made with the use of lactose (1%) instead of the blood. Media are adjusted to pH 7·2; the incubation temp. is 37°. P. S. ARUP.

**Utilisation of hop bitters in brewing.** P. Kolbach (*Mschr. Brauerei wissen. Beil.*, 1955, **8**, 119—125).—The influence of various factors on yields of bitter substances in the wort and beer is studied by means of experimental brewings. Duration of hop-boiling has a positive effect on the yields, but is probably not the sole factor; in the trials, pressure cooking offered a slight advantage. Increases in the amounts of bitter substances per boiling had less effect than might be expected, due to the more efficient utilisation of smaller proportions of hops. The degree of ageing of the hops (giving an increased % of hard resin on total resins) and low contents of coagulable N in wort used for hop-boiling had slight positive effects in the trials. Other factors examined are the pH of the wort and the beer, the amount of extract fermented, and the extent of multiplication of the yeast. Regarding the last two factors increases in the amount of $CO_2$ evolved probably increase losses of bitter substances. The effects of the above factors on foaming capacity and colloidal stability are discussed. P. S. ARUP.

**Determination of bitter-tasting conversion products of hops in worts and beer.** A. B. Moltke and M. Meilgaard (*Brauwelt*, 1955, **95, B**, 1265—1270).—The countercurrent extraction method of Rigby and Bethune for determining *iso*humulone, *iso*cohumulone, and *iso*adhumulone in wort or beer is examined, and minor modifications are recommended. A rapid simplified method proposed by Rigby (cf. J.S.F.A. Abstr., 1955, ii, 244) and improved by the author gives results agreeing (within 5%) with the results obtained by the above method, and showing reproducibility within 2%. In this method a single extraction with *iso*-octane (followed by centrifuging at 3000 r.p.m.) suffices, the total "*iso*humulone" content being determined spectrophotometrically in the *iso*-octane phase, on the basis of a mean extinction value at 275 mμ. for the three *iso*-humulones of 287. The whole operation must be carried out at 20° ± 0·5°. The method has proved reliable and suitable for routine work in determining the degree of utilisation of hop bitters, and their stability in beer; it is more reliable with respect to the bitters from fresh than from old hops. Methods are described for the prep. of an organoleptic "bitter scale" consisting in a series of beers of known *iso*humulone contents. P. S. ARUP.

**Modern views on beer proteins.** C. H. van den Noortgaete (*Fermentatio*, 1955, 143—159).—A review covering properties, classification, methods for separation, properties in relation to colloidal stability, and practical deductions. (49 references.) P. S. ARUP.

**Peptides and the melanoidin reaction.** H. Lüers and P. Lampl (*Brauwissenschaft*, 1955, **8**, 218—221).—At pH 5, glycylglycine and diglycylglycine both react with glucose at approx. six times the rate of the reaction between glycine and glucose; at pH 6, the corresponding ratios are 2·8 and 1·7, respectively. The difference is probably due to the greater distance between the COOH- and the $NH_2$-groups in the di- and tri-peptide. The small differences in reaction rates as between the di- and tri-peptide is probably due to space-relations connected with spiral structure. The importance of the lower peptides as sources of reducing agents and melanoidins in malting and wort-boiling is pointed out. P. S. ARUP.

**Colour and turbidity measurement of wort and beer by means of a new Zeiss electrophotometer. II.** H. Hecht (*Brauwissenschaft*, 1955, **8**, 207—214; cf. J.S.F.A. Abstr., 1955, ii, 289).—Tabulated transmission data for the Zeiss Elko II spectrophotometer, made at 350—750 mμ., with reference to the usual colour and turbidity standards, show that colour measurements of clear liquids can best be made at 380 or 420, and turbidity measurements at 720 or 750 mμ. The examination of coloured slightly turbid liquids can be based on measurements within two spectral ranges. Preliminary investigations of colour and turbidity determinations on worts are described. P. S. ARUP.

**Colour and turbidity measurements of wort and beer by means of a new Zeiss electrophotometer. III.** H. Hecht (*Brauwissenschaft*, 1955, **8**, 235—240; cf. preceding abstr.).—Turbidity measurements by means of the Elko II apparatus can be made at 750 mμ. without colour interference. A formula is given for correcting for turbidity colour measurements made at 380 mμ. Results of colour and turbidity measurements are tabulated for a large no. of worts and beers. Colour measurements should be made within 1 hr. after the wort is drawn off, in order to avoid complications due to darkening effects. The Elko II apparatus is suitable for use in the brewery laboratory; for routine purposes it could be simplified to give transmission % correct (at most) to the first decimal figure. Further simplifications are suggested which might reduce the cost of the apparatus. (33 references.) P. S. ARUP.

**Colloidal stability of beer.** J. Vandamme (*Fermentatio*, 1955, 175—189).—A review covering the physico-chemical properties of proteins with respect to colloidal properties, the effects of conditions during malting and brewing on colloidal stability, and the use of grains other than barley and diastatic prep. P. S. ARUP.

**Pasteurisation and pasteurising plant.** F. G. Redlbacher (*Brauer u. Mälzer*, 1955, **8**, No. 18, 3—7).—A review covering the construction and operation of modern pasteurising plant for bottled and bulk beer. P. S. ARUP.

**Bottling and canning of beer.** D. G. Ruff and K. Becker (*Brauer u. Mälzer*, 1955, **8**, No. 19, 8—16; in English pp. 12—16).—A review of the authors' book of the above title (Siebel Inst. Technology, Chicago). P. S. ARUP.

**Aëration as cause of inaccuracy in measurements of foam retention by beer.** W. J. Klopper (*Brauwelt*, 1955, **95, B**, 1097—1098).—The opinion of de Clerk and Bekisch (cf. *ibid.*, 640) that anomalous results obtained by the Ross and Clarke method are due to differences in the degree of aëration when the beer is poured into the cylinder is contested. The actual source of error is due to the fallacy of the assumption by Ross and Clarke that the course of the breakdown of the foam follows a linear pattern, whereas in fact, the relative stability of the remaining foam increases during the course of the breakdown. P. S. ARUP.

**Absorption of oxygen by beer in the bottle.** H. Graszmé and M. Sonntag (*Brauwelt*, 1955, **95, B**, 1413—1416).—Reference is made to the $O_2$ uptake during the drawing-off process, and experimental work is discussed mathematically on the uptake of $O_2$ in the bottle using filling tubes of various lengths, these being at different heights from the bottom of the bottle. The $O_2$ uptake in drawing off with a filling tube of average length was about 0·30 g./l. and increases with the shortening of the tube. With a tube 50 mm. long, the uptake is, at most, 0·15 mg./l. E. M. J.

**Biological method for detection of preservatives and antibiotics in beers.** R. Baetslé, D. A. A. Mossel and H. Verheyden (*Ann. Falsif., Paris*, 1955, **48**, 412—419).—In the test proposed by Mossel (cf. J.S.F.A. Abstr., 1955, i, 207) nutrients are added to beer before inoculation and incubation with yeast. It was found that greater sensitivity and a considerable shortening of the time required for test could be obtained by a preliminary incubation of the inoculated sample before addition of nutrients, the optimum period for such inoculation being 24 hr. A further method involving inoculation of separate samples with Gram-positive and -negative organisms, followed by acidimetry, enables specifically anti-bacterial antibiotics to be readily detected. (19 references.) J. S. C.

**Preservatives containing mercury, and their detection in beverages.** R. G. Eeckhaut (*Fermentatio*, 1955, 119—135).—The preservative sold under the names "Antibiotine Forta," "Détergent Sox," "F.A.," "Fongicine," etc. is found to be a solution containing $NaN_3$ and Na mercurihydroxybenzoate. The former substance is active mainly against Gram-negative organisms, whilst the latter inhibits bacterial activity by interfering with the enzymic functions of SH-groups. When added to beer in the proportion recommended, giving 0·7—0·8 p.p.m. of Hg, the prep. preserves the beer against light, but not against heavy infections of spoilage organisms. A method by which 0·15 p.p.m. of Hg can be detected in beer consists in the destruction of org. matter in the sample (500 ml. evaporated to 250 ml.) by means of $KClO_3$ + HCl + $CuSO_4$, pptn. of the Hg first as HgS, and then as Hg on fine brass wire from a solution of the HgS in HCl + $HNO_3$ which has been adjusted to pH 3, sublimation of the Hg from the wire on to a cooled portion of a test-tube, and finally rendering the Hg plainly visible as $HgI_2$ by exposure to I vapour. Various aspects of the subject, including the toxicity of Hg are discussed in detail. (32 references.) P. S. ARUP.

**Sorbic acid, a new preservative ; its detection in beverages.** R. G. Eeckhaut (*Fermentatio*, 1955, 136—142).—The prep. "Adox nouveau" is a solution containing per 100 ml., 12·5 g. of sorbic acid as the Na salt. The preservative properties of sorbic acid (in spite of its non-toxicity to animals) are confirmed, and theories as to the physiological mechanism of its bacteriostatic action are discussed. A method is described for the detection of sorbic acid which depends on its isolation (together with salicylic or other org. acids) by the usual methods, and on the fact that it yields a volatile reducing substance (acetaldehyde) on oxidation with $KMnO_4$ in acid solution. Sorbic and salicylic acids give the same coloration with $FeCl_3$, and both reduce $KMnO_4$, but the formation of acetaldehyde is specific for sorbic acid. The amounts detectable by this method are less than those required for effective preservation. (16 references.)
P. S. Arup.

**Sampling and process control in the dairy.** A. Eck (*Industr. aliment. agric.*, 1955, **72,** 495—498).—Problems of sampling and testing, in relation to process control and maintenance of required legal standards, in dairies are discussed on a mathematical statistical basis.
J. S. C.

**Physico-chemical properties of milk. V. Viscosity in milk.** B. R. Piri and H. L. Gupta (*Indian J. Dairy Sci.*, 1955, **8,** 78—82).—Milk stored for 10 hr. at 25°, for 6 hr. at 30° or for 3 hr. at 35° has a viscosity unchanged from that of the original. The proteins of milk make the greatest contribution to $\eta$ ; milk is a non-Newtonian liquid, i.e. its apparent $\eta$ varies slightly with rate of shear (flow). L. G. L. Unstead-Joss.

**Effect of antibiotics upon the microflora of milk.** R. Angelotti, H. H. Weiser, W. L. Slatter and I. A. Gould (*Appl. Microbiol.*, 1955, **3,** 234—237).—The effects of five antibiotics on the development in separated milk of test organisms are examined with a view to the possible control of lactic acid fermentations. Aureomycin, Terramycin and streptomycin are active against desirable and undesirable types of organisms, whilst penicillin and bacitracin are much less active against the latter than the former. P. S. Arup.

**Penicillin in milk.** J. C. Oosthuizen (*Fmg S. Afr.*, 1955, **30,** 267—268).—A microbiological test, based on the reduction in activity of *Strept. thermophilus* as measured by the amount of acid production, is described. E. G. Brickell.

**Simple method for determining the degree of neutralisation of milk.** K. Woidich and L. Schmid (*Z. LebensmittUntersuch.*, 1955, **102,** 167—171).—It is established by numerous tests that normal mixed milk (occurring, in practice, in bulks of >12 gal.) requires, irrespective of its initial pH or acidity, a constant vol. of standard acid for the reduction of its pH to 2·7. Any excess over this constant potentiometric titre, viz., 10·1 ± 0·1 ml. of 0·25N-HCl per 25 ml. of milk, indicates the previous addition of alkalinity to the milk. The presence in 25 ml. of milk of alkali derived from 0·0084 g. of $NaHCO_3$ can be detected by an excess in the titre of 0·4 ml. The titre of individual milks varies from 9·1 to 12·3, but that of a mixture of two such milks, even if both have abnormally high or low titres, approximates to the normal. Neither pasteurisation nor the presence of formaldehyde interferes with the test.
P. S. Arup.

**Circular paper chromatography method for the detection of adulteration in milk.** B. V. Ramachandra, N. N. Dastur and K. V. Giri (*Indian J. Dairy Sci.*, 1955, **8,** 83—88).—Milk adulterated by the addition of water with sufficient sucrose to make the solution isotopic with milk (as determined by f.p. tests) may be detected by a chromatographic technique, as little as 5% water (with the necessary sucrose) being detectable. The solvent is *n*-butanol–pyridine–water, and colour is developed by a mixture of 2% triphenyltetrazolium chloride and alcoholic KOH. L. G. L. Unstead-Joss.

**Abnormal freezing-point of milk.** A. Houlbrooke (*Chem. & Ind.*, 1955, No. 42, 1349—1350).—Instances are reported of milk found to have abnormally high f.p. (i.e. higher than −0·530°), the highest being −0·523°. Corresponding solids-not-fat and fat content figures are given, as are some particulars of feed, water supply and weather conditions. The high f.p. were found in the evening milk, and were usually associated with low f.p. in the morning milk.
J. S. C.

**Abnormally small freezing-point depressions of genuine milk.** D. J. T. Bagnall and A. Smith (*Analyst*, 1955, **90,** 623—625).—Samples of milk taken under supervision from a herd of 32 cows (mainly Friesians, 19 of which had calved within three months of sampling) gave average f.p. depressions as small as 0·518° and 0·522° (Hortvet) for the morning and evening yields respectively. The other analytical results suggest that these small depressions were due to deficiency of lactose caused by a recent change of diet from winter stall feeding to grazing on lush spring grass. This change probably caused a liberal intake of water to which the cows would not immediately adjust themselves. As the f.p. depression increased, the lactose and other constituents regained their normal values. Reports of abnormally small depressions, such as these, for genuine milk are very rare, and a depression of 0·530° (Hortvet) may still be generally regarded as the min. depression for such milk.
A. O. Jones.

**Cold sterilisation of liquid foods using mercury resonance radiation. I. Milk.** J. J. Albrecht, H. E. Gunning and M. E. Parker (*Food Res.*, 1955, **20,** 424—442).—General theories and mechanisms of resonance radiation are reviewed especially in regard to wavelengths <3000 A., dealing with proteins, enzymes, bacteria, off-flavours in irradiated milk, activated flavours, and whether milk could be subjected to resonance radiations under conditions that would provide the destruction of contained micro-organisms without encountering objectionable side-effects. Results indicate that this may be possible, and the contribution to the technological advance in the preservation of milk and its products is discussed. (50 references.)
E. M. J.

**Micrococci in milk. I. Incidence and distribution.** V. Lakshminarasim and K. K. Iya (*Indian J. Dairy Sci.*, 1955, **8,** 67—77).—Micrococci were found to constitute 67—77% of the total bacteria content and 9—20% of the micrococci were thermoduric. *Micrococcus caseolyticus, M. pyogenes albus, M. epidermidis, M. freudenreichii, M. candidus, M. conglomeratus, M. citreus, M. flavus, M. luteus, M. aurantiacus, M. roseus, M. rhodochrous* and *Sarcina lutea* were found. (35 references.) L. G. L. Unstead-Joss.

**Application of the coliform test to pasteurised milk and cream.** J. M. Frayer (*Vermont agric. Exp. Sta.*, 1955, *Bull.* 578, 14 pp.).—There was no relationship between coliform counts of raw milk and the same milk after pasteurisation. Organisms of the coliform group rarely survived pasteurisation in no. sufficient to be counted in 1-ml. samples. A standard of not more than 10 coliform bacteria per ml. for pasteurised milk and cream is considered fair and attainable. A. H. Cornfield.

**Determination of an inhibitory substance in milk—penicillin.** L. R. Mattick, E. O. Anderson and H. L. Wildasin (*J. Dairy Sci.*, 1955, **38,** 829—834).—A rapid method is described for the determination of low concn. (0—1·0 μg. per ml.) of penicillin in milk. The method is based on the inhibition of $NO_2'$ production from $NO_3'$ by *Micrococcus pyogenes* var. *aureus*. The decreased $NO_2'$ production in the presence of the antibiotic is measured by diazotisation of sulphanilic acid, coupling with α-naphthylamine hydrochloride and comparing the colour with that of controls. S. C. Jolly.

**Fat extraction from milker rubber with lye solutions.** J. M. Jensen (*J. Dairy Sci.*, 1955, **38,** 835—842).—Butter fat was effectively extracted from fat-saturated milking machine inflations by soaking for seven days in cold 2—15% aq. lye : concn. of 5—10% were most effective for latex-type rubber. Extraction for 60 min. with boiling aq. 10—20% lye was less effective, while lye solutions of the generally accepted concn. were almost useless. Butter-fat absorption and subsequent extraction were affected by rubber quality. Neoprene absorbed only small amounts of fat, which were best extracted with cold 2% lye. S. C. Jolly.

**Composition of the casein-containing particles in milk.** T. F. Ford, G. A. Ramsdell and S. G. Landsman (*J. Dairy Sci.*, 1955, **38,** 843—857).—Approx. 75—90% of the casein in fresh untreated skim milk exists as a single phosphoprotein (probably a mixture or combination of α-, β- and γ-casein in the proportion 16 : 4 : 1) which is contained in all colloidal particles, combined with Ca and $Ca_3(PO_4)_2$, in the intermediate size range. The proportion of inorg. P varies between milks and decreases with decreasing particle size, but with low-Ca milks and in the smallest particles a min. proportion of inorg. P occurs such that the mol. ratio, Ca : org. P : inorg. P is approx. 5 : 2 : 2. The largest casein-containing particles contain non-protein substances and less org. P. S. C. Jolly.

**Natural variation of milk serum-proteins as a limitation of their use in evaluating the heat treatment of milk.** H. A. Harland, S. T. Coulter and R. Jenness (*J. Dairy Sci.*, 1955, **38,** 858—869).—The variability in content of serum proteins in fresh milk and in their heat denaturability seriously limits the use of serum-protein analyses for assessing the previous heat treatment of milk powders of unknown history. The thiol content not only shows a natural variability but is affected by ageing of the milk and contamination with Cu. S. C. Jolly.

**Effect of artificial light on milk in cold storage.** A. C. Smith and P. MacLeod (*J. Dairy Sci.*, 1955, **38,** 870—874).—Ascorbic acid oxidation and oxidised flavour development were less and increased more slowly in pasteurised winter milk (homogenised and unhomogenised) stored in clear bottles at 45°F. in the dark than in milk exposed to 6, 12 and 18 ft.-candles of incandescent or fluorescent

light. Ascorbic acid retention was less in fluorescent light, but oxidised flavour development was similar with both types of light. S. C. JOLLY.

**Thiobarbituric acid test for milk lipin oxidation.** S. Patton and G. W. Kurtz (*J. Dairy Sci.*, 1955, **38**, 901).—Freshly prepared $\alpha\beta$-unsaturated aldehydes do not give the characteristic pink colour in the thiobarbituric acid test, but do so in presence of small concn. of $Cu^{++}$ or after autoxidisation for several days in air at 25°. The spectral characteristics of the colour developed by acrolein, crotonaldehyde and hept-2-enal, with absorption max. at 450 and 532 m$\mu$., are similar to those of the colour obtained in the test with oxidised lipin materials. S. C. JOLLY.

**Some factors affecting the quantity of water-insoluble fatty acids in cream.** L. K. Crowe (*J. Dairy Sci.*, 1955, **38**, 969—980).—Holding cream at low temp. was not effective in maintaining low values for water-insol. fatty acids (*A*); concn. of *A* decreased in some samples when held at 40—45° and 55—60°F. Acidification of cream with lactic acid generally tended to reduce levels of *A* on storage, but interfered with complete removal of *A* in Hillig's method of determination (*J. Assoc. off. agric. Chem.*, 1947, **30**, 575). Inoculation with starter had inconsistent effects on *A* concn. Excessive agitation of milk during cooling gave high concn. of *A*. Cream with high *A* contents can be detected by direct titration of centrifuged oiled-off fat from churned cream with alcoholic KOH. S. C. JOLLY.

**Preservation of Indian milk sweets.** *Shrikhand wadi* **and** *Milk burfee*. W. B. Date and D. S. Bhatia (*Indian J. Dairy Sci.*, 1955, **8**, 61—66).—*Shrikhand wadi* is prepared by souring milk with lactobacilli; the curds are mixed with an equal wt. of cane sugar and heated, with the addition of antioxidants. The cooled product is broken into pieces wrapped in waxed paper for storage. *Milk burfee* is prepared by adding *khoa* to hot syrup containing an equal wt. of cane sugar, and heating until a hard flat surface is obtained on spreading. Only *Shrikhand wadi* with a moisture content of 6·3% or less was acceptable after six months and the flavour of this sample had deteriorated due to the development of free fatty acids, which is not checked by antioxidant. Fatty acid contents (>~0·8%) were not acceptable. *Milk burfee*, however, kept well for six months, though samples with higher moisture levels (>9%) have a tendency to develop more peroxide, and antioxidant has a useful anti-rancidity effect. Packing in $N_2$ had no effect on keeping quality. L. G. L. UNSTEAD-JOSS.

**Pasteurisation equivalents of high-temperature short-time heating with ice-cream mix.** J. Tobias, O. W. Kaufmann and P. H. Tracy (*J. Dairy Sci.*, 1955, **38**, 959—968).—Using *Mycobacterium tuberculosis* as test organism, a temp. of 194·0°F. for 0·8 sec. (estimated holding time in Rosewell heater intended to have no holding time) or 186·0°F. for 3·8 sec. has a pasteurising effect on ice-cream mix equiv. to 175·0°F. for 25 sec. Corresponding temp. for 177·5°F. for 30 sec. (previously reported as equiv. to 160°F. for 30 min.) are 198·0° and 190·0°F., respectively. Using *Micrococcus* sp. MS 102, 187·2°F. for 0·8 sec. or 181·3°F. for 3·8 sec. is equiv. to laboratory pasteurisation at 155°F. for 30 min.; the corresponding temp. equiv. to 160°F. for 30 min. are 194° and 187°F., respectively. S. C. JOLLY.

**New application for the ultrasonic generator.** F. Rose (*Canad. chem. Process.* 1955, **39**, No. 11, 40).—The use of an ultrasonic generator for homogenising ice-cream ingredients is described. No pressure is necessary. The liquid raw material is sucked into the equipment, blown into a whistle by a pump and emerges at ~200 lb./sq. in. pressure from a slit-shaped nozzle. The flat jet produced impinges on a thin, sharp-edged, flexible, stainless-steel blade, clamped to vibrate at ~22,000 cycle/sec. Cavitation occurs with resulting pressures of up to 30,000 lb./sq. in., localised around the blade. The effect is being utilised in homogenising, emulsifying, dispersing, mixing, etc. The standard production unit handles 300—400 gal./hr. A positive-displacement gear pump will handle even thick slurries, but slow-speed gear pumps or progressive cavity-type pumps are used for highly viscous or abrasive liquids. Throughputs can be increased by connecting ultrasonic vibrator heads in parallel. O. M. WHITTON.

**[Determination of] foreign fats in dairy products. Critical review of the Reichert–Meissl and Polenske determinations.** S. D. Fine (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 319—338).—A glass-joint apparatus is described. For determination of Reichert–Meissl values, either this apparatus or that formerly used (*Analyst*, 1936, **61**, 404) may be employed, but for that of Polenske values the apparatus used must be specified. Gas-heating is recommended and carborundum is preferred to pumice as an anti-bumping agent. It is confirmed that varying pressure significantly affects the results. A. A. ELDRIDGE.

**Free amino-acids formed in various lactic acid starter cultures as measured by ion-exchange chromatography.** M. K. Hamdy, W. J. Harper and H. H. Weiser (*Appl. Microbiol.*, 1955, **3**, 221—226).—The proteolytic activity of starter-bacteria is shown by increases in amino-acid contents of the sera from ripened Italian separated-milk starters when these are stored at 10°. The occurrence of the amino-acids varies with the nature of the starter, but all the cultures (mixed strains of *Lactobacillus lactis*, *L. bulgaricus* or *Streptococcus thermophilus*) contain serine phosphate in notable concn. Serine phosphate (9 mg. per l.) was also found in (non-inoculated) separated milk after sterilisation and incubation; it increased in concn. to 40 mg. per l. during the first seven days of storage, after which there was a slow decrease to approx. the original value. The effect could not be traced to any viable organism. P. S. ARUP.

**Loss of vitamin A potency during the preparation of ghee from milk.** H. S. Patel and B. M. Patel (*Indian J. Dairy Sci.*, 1955, **8**, 53—60).—Milk from native cows was separated and the cream ripened for two days with a starter. The cream was directly boiled into ghee at a temp. >125°. A comparison experiment was performed, preparing butter from the cream before boiling. Ghee was prepared from buffalo milk by the indigenous method, curdling the milk, churning to form butter and boiling the butter. Vitamin A (**I**) was determined on the milk and ghee (after saponification by KOH) using the Carr–Price reaction. In the experiments with cows' milk, losses of **I** were ~30% and of carotene (**II**) ~21%. Ghee-making from colostral milk showed higher losses of **I** and **II** than from normal milk, as the initial concn. were very high. These losses are not influenced by the method of manufacture. Approx. 20% of **I** was lost when ghee was made from buffalo milk. L. G. L. UNSTEAD-JOSS.

**Identification of traces of isovaleric acid in butter sophisticated with hydrogenated whale oil.** G. D'Arrigo (*Olii min.*, 1955, **32**, 147—148).—An answer to criticisms. The author's method (cf. J.S.F.A. Abstr., 1955, i, 371) does not suffer from losses during the concentration of K salts, and the chromatogram spots are not diffuse. T. P. MCLAUGHLIN.

**Spectrophotometric detection of certain stabilisers in soft curd cheeses.** M. J. Gnagy (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 189—193).—To separate gums the cheese is treated with aq. $NH_3$ and then with acetic acid, the mixture is centrifuged and filtered, the filtrate being treated with alcohol and K alum solution. After decantation of the supernatant liquid, centrifuging and again decanting, treatment with alcohol, acetic acid and alum is repeated; finally the ppt. is dissolved in hot water containing $NH_3$ and re-pptd. as before. After drying and pulverising it is refluxed with dil. HCl and the resulting furfuraldehyde is distilled; its absorbence is then determined in a spectrophotometer at 277·5 m$\mu$. An absorbence greater than 0·25 indicates the presence of gum tragacanth, gum arabic, gum karaya, pectin, algin or propylene glycol alginate. A. A. ELDRIDGE.

**Modified Cornell phosphatase test for the analysis of blue mould and aged cheeses.** J. H. Mahon, C. Anglin and Ross A. Chapman (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 482—493).—The Cornell phosphatase test (Kosikowsky, *J. Dairy Sci.*, 1951, **34**, 1151) has been studied and an improved procedure is recommended. The slope of the phenol calibration line is depressed by all cheeses, and especially by blue mould and aged cheese; the effect is related to the free amino-acid content of the cheese. Higher phosphatase values are obtained by increasing the amount of 2 : 6-dichloroquinonechloroimide (CQC) reagent employed, and the use of a 1 : 6 dilution factor is recommended. The modified procedure is described. A. A. ELDRIDGE.

**Bacterial enzymic method for determining tyrosine in cheese.** G. J. Silverman and F. V. Kosikowski (*J. Dairy Sci.*, 1955, **38**, 941—949).—A rapid method is described for determining tyrosine in ripened Cheddar cheese based on conversion to tyramine with tyrosine decarboxylase and subsequent determination of the amine by a modification of the colorimetric method of Kosikowski and Dahlberg (*ibid.*, 1950, **33**, 438). S. C. JOLLY.

**Tyrosine in Cheddar cheese.** G. J. Silverman and F. V. Kosikowski (*J. Dairy Sci.*, 1955, **38**, 950—958).—The amount of free tyrosine (**I**) (348—2630 $\mu$g.) in well ripened commercial Cheddar cheese depends on differences between proteolysis and decarboxylation rates; total **I** (1341—2021 $\mu$g.) is a function of proteolytic enzyme activity, and may reach 3583 $\mu$g. per g. dry wt. in experimental cheeses. Addition of *Streptococcus fæcalis* to the starter does not affect total **I** liberation significantly, but higher amounts occur in experimental raw-milk cheeses than in those from pasteurised milk. Total **I** liberated is a more sensitive criterion of cheese ripening than are sol. protein values, and results apparently from hydrolysis of lower peptides. S. C. JOLLY.

**Chromatographic studies on proteolytic bacteria in their relationship to flavour development in Cheddar cheese.** A. R. Yates, O. R. Irvine and J. D. Cunningham (*Canad. J. agric. Sci.*, 1955, **35**, 337—343).—

Of 350 cultures tested, 24 were found capable of protein degradation at 13°. These included eight strains of *Streptococcus cremoris*, five of *S. lactis*, five of *Lactobacillus casei*, four of *S. liquefaciens*, one of *S. fæcalis* and one of *Sarcina lutea*. Undesirable flavours were produced by *S. liquefaciens* cultures only. After 3 to 10 months' curing, vats containing *L. casei* and *Sarcina lutea* had more flavour and better texture than the controls. E. G. BRICKELL.

**Microbiology of the surface ripening of brick cheese.** D. J. Lubert and W. C. Frazier (*J. Dairy Sci.*, 1955, **38**, 981—990).—Micrococci are the chief organisms concerned in odour and flavour production in brick cheese. Flavour is intensified by the previous growth of film yeasts in the smear that develops on the surface early in the ripening. The predominant micrococci are colourless variants of *M. varians*, but *M. caseolyticus* and *M. freudenreichii* are more active flavour producers. Acetic, butyric and sometimes propionic acids contribute to flavour, but higher volatile fatty acids are responsible for the sweaty odour of the smear. S. C. JOLLY.

**Salting-out curve of egg-white proteins.** J. C. Perrone, D. M. Peixoto and E. Tolmasquim (*An. Acad. brasil. Cienc.*, 1955, **27**, 167—168).—Egg-white proteins were fractionated by salting out with phosphate solutions (prepared according to A. M. Butler and H. Montgomery, *J. biol. Chem.*, 1932, **99**, 173) of concn. rising by steps of 0·545M. from 1·091M. to 2·727M. The procedure adopted was to add 10 ml. of solution to 1 ml. of filtered, undiluted egg-white, filter through Whatman 42 paper until perfectly clear, dilute 1 : 10 with 0·1N-NaOH, and take readings at 280 mμ. in a Beckman DU Quartz spectrophotometer. The curves showed the existence of twelve fractions (eight globulins and four albumins); most were not identifiable with known protein fractions obtained by other methods, e.g., electrophoresis, but the third fraction was tentatively identified as ovalbumin, representing ~60% of total proteins, and the eighth appeared to correspond with the $G_1$ fraction obtained by electrophoresis. J. S. C.

**Physical and functional properties of lyophilised whole egg, yolk and white.** T. Rolfes, P. Clements and A. R. Winter (*Food Technol.*, 1955, **9**, 569—572).—Lyophilisation had no detrimental effect on the functional properties of albumin as measured by angel cake volume. Lyophilisation of yolk impaired its emulsifying properties as measured by mayonnaise stability; and of whole egg impaired its functional properties as measured by sponge cake volume. (15 references.) E. M. J.

**Photoelectric inspector detects green rot in eggs.** K. H. Norris (*Electronics*, 1955, **28**, 140—142).—U.v. light detected by two multiplier phototubes measures fluorescence of bacterial spoilage inside eggs at rates up to 500 eggs per min. Use of response ratio at two wavelengths makes measurements independent of *d* and colour of the shell. SCI. ABSTR. (R. B. C.).

**Evaluation of egg colour.** W. D. Pohle and V. C. Mehlenbacher (*Food Technol.*, 1955, **9**, 565—568).—The use of pure carotene as a standard substance and the reporting of egg colour in terms of carotene is proposed, as a means of overcoming variation in values for egg colour measured at different wavelengths and with different instruments. The colour is measured in the region of 455 mμ. and the results are expressed on a sample wt. basis. E. M. J.

**Growth of bacteria on horse muscle, in relation to the changes after death leading to *rigor mortis*.** M. Ingram and G. C. Ingram (*J. Sci. Food Agric.*, 1955, **6**, 602—611).—The intrinsic *Clostridia* in the muscles of whale carcasses multiply appreciably after the muscles have passed into *rigor mortis*, but not before this stage. Attempts to detect a similar phenomenon in the *sterno-cephalicus* muscles of horse carcasses failed to show any delay in bacterial growth on pre-rigor as compared with post-rigor muscles. The implications of this difference are discussed. J. S. C.

**Heat processing of beef. IV. Functional relationships of temperature, time and space during processing at high retort temperatures. V. Temperature distribution patterns during processing of beef at high retort temperatures.** H. Hurwicz and R. G. Tischer (*Food Res.*, 1955, **20**, 377—398, 399—414).—IV. Thermal properties of round of beef (thermal diffusivity, characteristics of heating and cooling curves) and temp. distribution during processing, over a wide range of temp. to include temp. near 300°F.; heating and cooling process and empirical relationships among variables encountered, etc., were studied to establish wherever possible the fundamental laws in mathematical form. The resulting equations are obtained by rigorous statistical methods and carry an experimental error estimate. (17 references.)

V. Experimental temp. distributions during processing of round of beef at six retort temp. ranging from 225 to 315°F. were determined, and found to be in disagreement with theoretical expectations. These data are discussed. E. M. J.

**Effect of maize, barley, stilbœstrol and degree of finish on quality of beef.** M. Simone, F. Carroll, E. Hinreiner and M. T. Clegg (*Food Res.*, 1955, **20**, 521—529).—In tests on four groups of steers (*a*) barley-fed to U.S.A. Dep. Agric. good grade; (*b*) barley-fed to choice grade; (*c*) barley-fed to choice grade and implanted with stilbœstrol (60 mg.) in the ear of each steer; (*d*) maize-fed to choice grade, results indicated that there were few significant differences among the four groups in the quality factors judged, e.g., tenderness, juiciness, flavour due to degree of finish, implantation with stilbœstrol, or the particular grain used for fattening. (19 references.) E. M. J.

**Interactions between ascorbic acid and psychrophilic bacteria associated with the discoloration of prepackaged beef.** R. N. Costilow, B. A. Batshon, L. J. Bratzler and D. A. Robach (*Food Technol.*, 1955, **9**, 560—563).—Ascorbic acid, 0·1% concentration, had an inhibitory effect on two strains of *Pseudomonas* spp. tested in nutrient broth and at the same time oxidation of the ascorbic acid was protected; concentration of total vitamin C-like compounds was not influenced. A desirable colour could be maintained longer in fresh beef steaks by keeping the initial bacterial contamination low than by treatment of highly contaminated steaks with ascorbic acid. (10 references.) E. M. J.

**Utilisation of sugar in meat processing.** R. Grau (*Fleischwirtschaft*, 1955, **7**, 182—184).—The advantageous effects of adding a small amount of sugar (0·2—0·5% on meat) during meat curing processes and dried sausage processing are discussed. It is suggested that bacterial processes are involved, resulting in improvements in flavour, tenderness, colour change, etc., and that the sugar contributes to the maintenance of the optimum pH.
SUG. IND. ABSTR. (E. M. J.).

**Effect of storage on the composition and nutritive properties of farm-style hams.** M. D. Fields, C. F. Dunker and C. E. Swift (*Food Technol.*, 1955, **9**, 491—495).—The hams were dry cured with salt only, smoked and stored at 40°, 70° and 90°F. for six weeks, and six- and 12-month periods. An average wt. loss of 11·5% occurred during processing; the salt content varied widely; the content of protein, fat, ash and NaCl increased as the hams became dehydrated; fat hydrolysis occurred in those stored for 12 months at 70° and 90°F.; no deterioration of riboflavin or niacin occurred, but there was reduction in the thiamine content. Hams deteriorated appreciably on storage for six months at 70°F., those at 90°F. were scored undesirable after storage for six months. Bacterial contamination was not the decisive factor in determining the edible storage life of the hams. (14 references.) E. M. J.

**Importance of bacteriological condition of minor ingredients in canned meat products.** D. A. A. Mossel (*Ann. Inst. Pasteur, Lille*, 1955, **7**, 171—179).—A review covering possible sources of infection, especially of pasteurised canned ham (>3 lb. in wt.), by sporing bacteria (viz. in spices, salts, sugars, starches, milk powder, new cans, and water) and appropriate counter-measures. (95 references.) P. S. ARUP.

**Determination of starch in meat products with the anthrone reagent.** F. J. Stevens and R. A. Chapman (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 202—210).—The sample, mixed with water, is stirred after addition of Zn acetate and K ferrocyanide, and the mixture is centrifuged. The residue is heated with dil. $H_2SO_4$, cooled, filtered, diluted to a definite vol., and after addition of the anthrone reagent the absorbence is measured at 620 mμ. Recoveries of added starch were ~95%. Glycogen or dextrose present in liver interferes. The method does not always give consistent results. A. A. ELDRIDGE.

**Effect of irradiation on meat fats.** M. Sribney, U. J. Lewis and B. S. Schweigert (*J. agric. Food Chem.*, 1955, **3**, 958—960).—A study of irradiation effects on meat fats indicated that fat is not the constituent in meat responsible for off odours and flavours observed during irradiation. On subsequent storage, increases in peroxides, carbonyl compounds, and free fatty acids were small when the presence of $O_2$ is minimised. There was a marked increase in peroxide values when irradiated fats were stored at 5° in an $O_2$-permeable casing as compared with non-irradiated fats stored in similar casing. (11 references.) E. M. J.

**Manufacture of gelatin.** A. Rousselot (*Chim. et Industr.*, 1955, **74**, 669—680).—The manufacture of gelatin from the bones and skins of animals, methods of test, and industrial applications are described briefly. J. M. JACOBS.

**Extracts and enzymic hydrolysates from fish liver and mammalian liver.** A. Guttmann (*J. Fish Res. Bd Can.*, 1955, **12**, 637—645).—Extracts and enzymic hydrolysates were prepared from livers of cod, haddock, salmon, whale, seal, and beef and pork livers, by extraction with water at pH 5, enzymic hydrolysis with papain at pH 5·5 and 65°, followed by pancreatin at pH 7·5 and 50°, after

which both extracts and hydrolysates were concentrated by distillation *in vacuo* to a syrupy liquor and then dried *in vacuo* at low temp. The resulting material was analysed in respect of: water, ash, Ca, P, Cu, Fe, Co, F, total $N_2$, amine-$N_2$, choline, thiamine, riboflavin, folic acid, niacin, pantothenic acid and vitamin $B_{12}$. The results are tabulated and discussed from the point of view of utilisation of fish liver extracts and hydrolysates in nutrition and medicine. (16 references.) J. S. C.

**Recent developments in the freezing of fish at sea. II. Quality of sea-frozen cod.** A. Banks (*Chem. & Ind.*, 1955, No. 43, 1360—1362).—Empirical investigations on the quality of cod treated in different ways before freezing and cold storage have shown that the fish should be gutted and well-iced very soon after catching, that they should not be held for longer than three days in ice before freezing, and that the frozen products should be cold-stored at −28° to −30°. J. S. C.

**Conversion of fish stickwater (press-water) to solubles. I. Bacterial decomposition of stickwater at high temperatures.** R. A. Macleod, D. R. Idler and W. A. B. Thompson. **II. Prevention of bacterial decomposition of stickwater at high temperatures.** D. R. Idler, R. A. Macleod and W. A. B. Thomson (*Appl. Microbiol.*, 1955, **3**, 202—204, 205—208).—I. The loss of solids (15—20%) occurring in herring press-water when spontaneously cooling from ~93° to 60° (in preparation for treatment with proteolytic enzymes) is shown to be due to aërobic and anaërobic bacterial action, which is most active at 60—65°. The mildest heat treatment capable of preventing bacterial action is autoclaving at 120° for 30 min.

II. Loss of solids by bacterial action in press-water at 60° can be economically and completely prevented during 48 hr. by the addition of 2 p.p.m. of penicillin *G*. At lower temp. (40—55°), at which losses are much greater, 1 p.p.m. is effective. Penicillin *G* at 2 p.p.m. retains its efficiency during 10 hr. in press-water at 71·3°, but tends to deteriorate at 82·2°. Aëration of the hot liquid causes appreciable loss of penicillin. P. S. Arup.

**Expressible fluid of fish fillets. IV. Iced cod.** A. Banks (*J. Sci. Food Agric.*, 1955, **6**, 584—587).—The amount and composition of expressible fluid from gutted cod iced for various periods were studied. The amount of fluid increased rapidly as the fish passed out of *rigor mortis*, then only slowly for a period, and then fairly rapidly again after 168 hours on ice. It is suggested that the results obtained are associated with physical changes in texture during gradual resolution of *rigor mortis* and with subsequent slight changes in texture and in the colloidal proteins of the protoplasm. The size of fillets appears to be unrelated to the amount of fluid. J. S. C.

**Retaining the quality of fishery products.** C. Butler (*Industr. Refrig.*, 1955, **129**, No. 3, 18—19).—Bacterial spoilage, development of rancidity in fish of high oil content, oxidation of unsaturated fats and oils in fish, and protein denaturation in fish, are discussed and recommendations are made in respect of freezing procedures, handling, processing, transport and related processes. J. S. C.

**Rapid determination of moisture content in fish meat.** Y. Tsuchiya and T. Nakaro (*Tohoku J. agric. Res.*, 1954, **5**, 93—97).—The method is based on stirring the chopped fish with methanol and calculating the water present, after 30 min., in the alcohol from the sp. gr. A correction factor which differs for each species of fish, is utilised. The average error of the method ranges from 0·50 to 1·71% according to species. A. G. Pollard.

**Thermodynamic properties of fish and their effect on the rate of freezing.** R. A. K. Long (*J. Sci. Food Agric.*, 1955, **6**, 621—633).—The variations with temp., which are considerable, of thermal conductivity, density, and apparent sp. heat, of fish muscle, are examined theoretically, and experimentally by the freezing of small packages of fillets by refrigerated air blast. The initial loading temp. was found to have a significant effect on the thermal arrest time and this is examined in detail. The effect is regarded as due to a combination of factors and further work on a quant. basis is envisaged. J. S. C.

**Transport of frozen fish.** E. Waller (*World Refrig.*, 1955, **6**, 499—502).—A review of distribution in Great Britain with tabulated data concerning the storage life of various types of fish. J. S. C.

**Component fatty acids and unsaponifiable compounds in shark liver oil.** G. G. Kamath and N. G. Magar (*J. Indian chem. Soc.*, 1955, **32**, 455—462).—Waghbeer and Khada mushi liver oils are studied for their component fatty acids by low-temp. separation, Me ester fractionation and alkali isomerisation. In Waghbeer liver oil, $C_{20}$- and $C_{22}$-acids predominate over the $C_{18}$- and $C_{16}$-acids; in Khada mushi liver, $C_{18}$-acids form the major portion. The unsaponifiable compounds of both oils are studied chromatographically and spectrophotometrically. Cholesterol, vitamin A and kitol are present in both samples. Full details of results are given, including photometric data. C. A. Finch.

**Isolation of *n*-pentadecoic and *n*-heptadecoic acids from shark *Galeorhinus australis* (Macleay) liver oil.** I. M. Morice and F. B. Shorland (*Biochem. J.*, 1955, **61**, 453—456).—*n*-Pentadecoic and *n*-heptadecoic acids are isolated from liver oil of the New Zealand school shark (by methods which involve fractional distillation of esters and crystallisation of the acids, without hydrogenation) in amounts corresponding with 0·28 and 0·17% of the total fatty acids, respectively. J. N. Ashley.

**Fluorimetry of oils.** A. Arpino, G. Ricca and G. Jacini (*Olii min.*, 1955, **32**, 149—153).—Spectrophotometric measurements of the fluorescence of 50 samples of olive oil, unaltered and decolorised with Norit, show peaks of emission at 610 mμ. and 504—587 mμ. Decolorised samples show a strong emission at 438—471 mμ. which is much weaker in untreated samples: addition of chlorophyll extracted from olive leaves to the decolorised oil has a similar inhibiting effect on this emission. Fluorescence figures are tabulated for all the samples, together with analytical data and refractive indices. T. P. McLaughlin.

**Antioxidant properties of spices in oil-in-water emulsions.** J. R. Chipault, G. R. Mizuno and W. O. Lundberg (*Food Res.*, 1955, **20**, 443—448).—Of 32 varieties of spices tested by the active $O_2$ method at 98·6°, all exhibited antioxidant properties in lard (*ibid.*, 1952, **17**, 46). The antioxidant effectiveness of spices in an oil-in-water emulsion, representing the basic conditions in food products in which the fat is in contact with an aq. phase, was studied. All spices protected the emulsion against $O_2$ absorption, cloves being very effective. Other spices with antioxidant indexes >5·0 were allspice, cardamon, cassia, cinnamon, ginger, mace, etc. E. M. J.

**Spectrophotometric method for determining piperine in oleoresins of black pepper.** H. J. Fagen, E. P. Kolen and R. V. Hussong (*J. agric. Food Chem.*, 1955, **3**, 860—862).—The chemical method for determining the piperine content of oleoresins of black pepper is based on the determination of total N by the Kjeldahl–Wilfarth–Gunning method, but this may give varied results caused by the presence of other nitrogenous substances. A spectrophotometric method is described for the measurement of the u.v. absorption spectrum of an oleoresin of black pepper in chloroform at 345 mμ. The recovery of pure piperine in oleoresins ranged from 96·7 to 102·9%. E. M. J.

**Sulphate–nitrate formation in semi-micro analysis.** J. Pien (*Ann. Falsif. Paris*, 1955, **48**, 420—421).—Results of semi-micro techniques of salt formation with $H_2SO_4$ and $HNO_3$, of samples of milk powder, butter and flour are reported using the apparatus previously described by the author (cf. J.S.F.A. Abstr., 1955, i, 277), with 1-g. as compared with 10-g. samples, in the same apparatus and by the classical open flask method. In all cases, a very substantial reduction in the time required for the process was obtained. J. S. C.

**Size and form of the molecules in γ-globulin.** O. Kratky and B. Paletta (*Angew. Chem.*, 1955, **67**, 602—603).—Use of the small-angle *X*-ray method has shown that the γ-globulin mol. possesses the form of an elliptical cylinder. For the first time all three axial values of a dissolved corpuscular albuminous substance are determined by direct measurement. The theoretical background of these studies is outlined. C. A. Finch.

**Paper chromatography in routine determination of glutamic acid in production.** N. A. Khan, B. E. Baker and W. F. Van Horn (*J. agric. Food Chem.*, 1955, **3**, 853—855).—In the process described, the glutamic acid is determined on buffered paper (pH 12) using a solvent system made up of buffer (pH 12) saturated phenol containing a small amount of 2-butanol, the use of this solvent system resulting in a clean separation of glutamic acid from the other amino-acids, in round, compact spots. E. M. J.

**Cystine, tyrosine and essential amino-acid contents of selected foods.** C. H. Edwards, L. P. Carter and C. E. Outland (*J. agric. Food Chem.*, 1955, **3**, 952—957).—Concentrations of ten essential amino-acids were determined in 25 selected foods rated as excellent to fair to poor sources of the acids, the data being presented and discussed. Possible supplementary relationships of the amino-acids in dietary planning are considered. (24 references.) E. M. J.

**New method for direct isolation of glycine from protein hydrolysates.** A. S. M. Selim, M. E. A. El-Wahab and M. M. El-Sadr (*Biochem. J.*, 1955, **61**, 177—179).—Glycine alone, of the amino-acids normally occurring in a protein hydrolysate, forms a sparingly-sol. complex salt with Cu picrate, in the mol. ratio 2 : 1. This forms the basis of a new method of isolating glycine, which is simple and economical in time and reagents. The complex (representing up

to 98·5% recovery of glycine) is readily decomposed by acids to give ~90% recovery of the protein. The introduction of undesirable reagents is avoided, and, after removal of glycine, the hydrolysate is in a form suitable for isolation of other amino-acids. Full details of the procedure (a simple pptn.) are given; the initial ppt. should be washed and recrystallised. With proteins of small glycine content, e.g., egg albumin, $NH_3$ should first be removed and a considerable excess of Cu picrate used; the amount of complex represents ~90% recovery of glycine. K. E. J.

**Utility of the "albuminoid ammonia value" in the analysis of foodstuffs. I. Analysis of vinegar.** S. N. Mitra (*J. Indian chem. Soc., Industr. Edn.*, 1955, **18,** 119—121).—Estimation of the albuminoid $NH_3$ affords a rapid sorting test for detecting whether malt vinegar has been sophisticated with artificial vinegar; determination of total N by the time-consuming Kjeldahl technique (hitherto a routine test) can be dispensed with. The total N content, and hence the total protein content, can be calculated approx. from the albuminoid N. G. HELMS.

**Separation of peptides from salt solutions by an ion-exchange method.** N. Haugaard and E. S. Haugaard (*C. R. Lab. Carlsberg, Sér. chim.*, 1955, **29,** 347—349).—A method is given for the separation of amino-acids and peptides from salt solutions using a cation-exchange resin. Peptides in quantities of <1 μ. mol. can be recovered free of salt from 0·1—0·2 M-citrate, phosphate or bicarbonate buffers having a vol. of ~100 ml., by adsorption on Dowex-50 in the $NH_4$ form, followed by washing with dil. acetic acid and elution with dil. aq. $NH_3$. I. JONES.

**Polarographic determination of the riboflavin and thiamine content of foods.** E. Kervei, M. Kiszel and M. Simek (*Acta chim. hung.*, 1955, **6,** 345—363).—A description is given of a method for the polarographic determination of riboflavin (**I**) and thiamine (**II**) in foods, which is based on the reduction of the vitamins on the dropping Hg cathode. Current–voltage curves are prepared from polarography of solutions of known content (100—160 μg.) of each of the vitamins, buffered by a phosphate buffer of pH 7·6 in the case of **I** and by an acetate buffer of pH 6·2 in the case of **II**; this known amount of **I** or **II** is then added to the solution to be estimated and the polarograph of the mixed solution is taken. The solutions of necessary purity and concn. for polarography are made by (*a*) extracting the vitamins from the foods (yeasts, liver, spinach, etc.), with 0·1N-HCl solution for **I**, and with 0·1N-HCl and 80% EtOH for **II**, and (*b*) enriching the resulting solutions and removing interfering substances by absorbing the vitamins (on fuller's earth activated with KCl for **I** and with active C for **II**), washing out the interfering substances from the adsorbents, and eluting the vitamin from the adsorbent (with hot 80% acetone for **I**, and with HCl–EtOH for **II**, the elution of **I** being quant., but of **II** yielding up to 90% only). The solution obtained is used for determining the amount of free vitamin present. For finding the total free and combined vitamin present in the sample, and hence enabling the amount of bound vitamin to be determined, part of the solution resulting from step (*a*) is subjected to the action of taka-diastase to free the bound vitamin before the enriching and subsequent steps. Full details are given of the procedures for various foods. In the estimation of **I** the results are accurate within 4—6%, provided that the content is ⩾400 μg. per kg. H. L. WHITEHEAD.

**The interplay of science and human values in food.** C. G. King (*Food Technol.*, 1955, **9,** 483—486).—An address dealing with development in food production, processing and distribution resources and the effects on human health, and a discussion on vitamin-C research. E. M. J.

**Manometric determination of cocarboxylase.** V. Boffi, A. Lucarelli and E. Bucci (*Ric. sci.*, 1955, **25,** 2069—2076).—Manometric determination of cocarboxylase (DPT) in pure solution and in biological materials is rendered more accurate by using a pure solution of apocarboxylase as coenzyme instead of yeast extracts containing phosphatases which destroy DPT. T. P. MCLAUGHLIN.

**Chromatographic separation of adenosine triphosphate [ATP] and its organic and inorganic derivatives.** P. Cerletti and N. Siliprandi (*Ric. sci.*, 1955, **25,** 2084—2090).—A paper chromatographic method is described for separating adenosine and inosine di- and tri-phosphates, muscle adenylic acid, adenosine, adenine, pyrophosphate and orthophosphate, using as eluant 75 : 20 : 5 : 0·3 propanol–water–trichloroacetic acid–22°Bé. aq. $NH_3$. The ATP deriv. are viewed by u.v. light; the inorg. phosphates by molybdic acid development. T. P. MCLAUGHLIN.

**Absorption curves of Pyronines and their tinctorial powers in ribonucleic acid identification.** F. M. Gerola and M. Grilli (*Ann. Fac. Agr. Univ. cattol. S. Cuore*, 1955, **52,** Ser. ii, 152—160).—There are marked differences in spectrophotometric behaviour of a no. of commercial samples of Pyronine, and these differences are suggested as related to unsatisfactory behaviour in their use for cytochemical localisation of ribonucleic acid. F. R. PAULSEN.

**Technique for direct determination of the equilibrium relative humidity of foods.** D. A. A. Mossel and H. J. L. van Kuijk (*Food Res.*, 1955, **20,** 415—423).—A LiCl-cell is described and illustrated for the determination of the equilibrium relative humidity (*h*) of foods, i.e., the degree of desiccation or the concentration of solutes. Limiting values of *h* are given for common bacteria, yeasts and moulds occurring in foods. The values obtained by this method are in good agreement with data obtained by the classical, static method, but require much less time, one hour at most compared with at least one day. (49 references.) E. M. J.

**Aluminium with food and chemicals.** Northern Aluminium Co., Ltd. (1955, 90 pp.).—The properties of Al and its principal alloys, with particular reference to corrosion resistance, are described; the various types of attack are classified; and methods of avoidance, reduction or control of attack and of surface treatment of Al are listed. An alphabetical list of over 500 common foods or chemicals forms the main section of the pamphlet, and describes the reactions between each substance and Al and, where appropriate, the uses of Al or its alloys in connexion with it. J. S. C.

**Separation of permitted coal-tar food colours by paper chromatography. I.** S. N. Mitra and R. K. Chatterji (*J. Instn. Chem. India*, 1955, **27,** 169—176).—The coal-tar colours (water-sol. acid dyes, viz. tartrazine, indigotine, amaranth, Orange I and erythrosine) whose use in foodstuffs is permitted are separated and identified both in aq. solution and in one article of food (fruit squash) by paper chromatography. Two techniques are used: (*a*) vertical-column paper chromatography (ascending type) and (*b*) circular-paper-disc (horizontal type). The most suitable solvents are (i) 5% NaCl solution, and (ii) 1·0—0·25N-HCl. The order of separation (in decreasing height) is as given above. I. JONES.

**Colorimetric determination of vanillin.** D. T. Englis, J. W. Miles and L. A. Wollerman (*J. Ass. off. agric. Chem. Wash.*, 1955, **38,** 519—523).—The formation of a ppt. on addition of the $Na_2CO_3$ solution in the Folin–Denis method for the colorimetric determination of vanillin can be prevented by addition also of a small quantity of Na hexametaphosphate. Slightly higher absorbencies are then observed. A. A. ELDRIDGE.

**Method of choosing judges for a sensory experiment.** C. Y. Kramer (*Food Res.*, 1955, **20,** 492—496).—Reference is made to D. W. Chapman, the statistics and methods of correct matchings, *Am. J. Psychol.*, 1934, **46,** 287—298. E. M. J.

**Significance of taste.** E. Abderhalden (*Schweiz. Brauerei Rdsch.*, 1955, **66,** 164—166).—A review covering physiological aspects of the sense of taste. P. S. ARUP.

**Development of a scale for measuring soldiers' food preferences.** Lyle V. Jones, D. R. Peryam and L. L. Thurstone (*Food Res.*, 1955, **20,** 512—520). E. M. J.

**Single-sample method for foreign flavour detection.** A. M. Neubert and G. H. Carter (*Food Technol.*, 1955, **9,** 572—575).—A single-sample task method is described and taste-panel evaluation of insecticide-induced foreign flavours (apple juice and apples) is discussed. Apples treated with Demeton and juice prepared from such apples have a detectable foreign flavour. (12 references.) E. M. J.

**Food perishability in terms of micro-ecology.** D. H. F. Clayson (*J. Sci. Food Agric.*, 1955, **6,** 565—574).—The following factors involved in microbial spoilage of foods are discussed: chemical composition (including pH effects), infection, equilibrium humidity, storage temp., $O_2$ partial pressure, synergism and antagonism between micro-organisms. (50 references.) J. S. C.

**Vacuum contact-plate dehydration of foodstuffs. I. First appraisal.** E. G. B. Gooding and E. J. Rolfe (*J. Sci. Food Agric.*, 1955, **6,** 427—433).—Factors affecting the drying of foodstuffs and two methods of drying, the hot-air drying working on the conventional principle, and the vacuum contact-plate process are discussed. Of the two the vacuum contact-plate dehydration process possesses substantial advantages when considered in relation to the "ideal" dehydrated foods. Relevant factors are the absence of $O_2$ and the milder heat treatment of the foodstuff. The considerable cost of the equipment could be minimised by using the plant throughout the year to dehydrate any commodity in season. E. M. J.

**Vacuum contact-plate dehydration of foodstuffs. II. Equipment.** J. M. Hay (*J. Sci. Food Agric.*, 1955, **6,** 433—440).—Further consideration is given to the vacuum contact-plate dehydration process (cf. preceding abstr.). The equipment comprising a large steel vacuum cabinet with 21 trays holding about a ton of material to

be dried per batch, by 24 heating plates, is described in detail and illustrated. Good thermal contact is maintained with the foodstuff by the movable heating plates, resulting in the successful drying of meat steaks, fish fillets, sliced root vegetables, and certain whole fruits. E. M. J.

**Microbiological spoilage of canned vegetables and fruits. ii.** W. J. Hoppenbrouwers (*Conserva*, 1955, **4**, 7—12; cf. *ibid.*, 1955, **3**, 362).—A review covering spoilage by mesophilic and acid-resisting bacteria, yeasts, and moulds. (46 references.) P. S. Arup.

**Preservation of foods. VII. Preservation by heating. v.** J. A. Glerum and H. A. Leniger (*Conserva*, 1955, **4**, 13—20; cf. *ibid.*, 1955, **3**, 377).—A review covering practical directions. P. S. Arup.

**Preservation of foods. VIII. Preservation by refrigeration. i.** D. A. A. Mossel (*Conserva*, 1955, **4**, 115—122; cf *ibid.*, 13, 49, 85).—A review covering effects of refrigeration on foods, micro-organisms and pests, and practical expedients for increasing efficiency and hygiene in the cold store. P. S. Arup.

**Refrigeration equipment in the field of agriculture and food in France, 1955.** Section Technique du Froid du Ministère de l'Agriculture (*Rev. gén. Froid*, 1955, **32**, 1029—1036).—A general and statistical review of the plant and industrial applications of refrigeration and cold storage in the industries concerned with ice manufacture, meat storage, milk and dairy products, ice-cream, eggs and poultry, fruit and vegetables, and wines. J. S. C.

**More miles per pound of food.** A. C. Avery (*Food Technol.*, 1955, **9**, 533—535).—The preparation and packaging of food stores for ships, particularly submarines, are discussed. E. M. J.

**Protective packaging of foods against moisture condensation.** J. G. Woodroof and E. K. Heaton (*Food Technol.*, 1955, **9**, 510—518).—The nature of moisture condensation and measures to control condensation by protective packaging were determined on tin cans, fibreboard containers, glass jars, flexible containers, and certain products, e.g., nuts, dried fruits, etc. Canned vegetables were packed in cartons constructed in 26 ways, and removed from 32°F. and 75% R.H. to 70°F. and 95% R.H. Best protection was obtained by the use of cartons with Al foil on the inside and outside surfaces with corrugated pads on top and bottom of the cans, the cartons being closed by glue + the use of six strips of waterproof tape. (18 references.) E. M. J.

**Extension of food storage life by irradiation.** B. E. Proctor, J. T. R. Nickerson, J. J. Licciardello, S. A. Goldblith and E. E. Lockhart (*Food Technol.*, 1955, **9**, 523—527).—Tests were made to find whether certain foods could be irradiated at levels sufficient to destroy psychrophilic bacteria without causing unacceptable flavour changes. In hermetically sealed containers the storage life could be extended of (*a*) spinach held at 36—40°F. after treating with high-voltage cathode rays at a dose level of $1{\cdot}5 \times 10^6$ rep.; (*b*) fresh pork sausage links; (*c*) sausage patties and (*d*) ground beef each at a dose level of $1{\cdot}0 \times 10^6$ rep. Deterioration through microbial action of all-beef frankfurters could be delayed to some extent by irradiation at dose levels as low as $0{\cdot}25 \times 10^6$ rep. Off-flavour in beef was obviated by adding 0·3% of sodium fumarate and 0·5% of monosodium glutamate. E. M. J.

**Influences of ionising radiations on the protein components of selected foods.** F. J. McArdle and N. W. Desrosier (*Food Technol.*, 1955, **9**, 527—532).—Influences of ionising radiations on purified casein and egg albumin when these proteins are irradiated in aq. solution were studied. With casein there was a splitting of the mol. into smaller fractions, then association of these fractions until complete coagulation occurred. The mol. of egg albumin were transformed from a globular form to an asymmetrical mol.; association of the transformed mol. resulted in coagulation. Trypsin hydrolysis rates of casein and egg albumin were increased. All these effects were reduced when the proteins were irradiated in the presence of ascorbic acid. The effects of cathode rays on the electrophoresis pattern and mobilities of casein and of egg albumin are discussed. (15 references.) E. M. J.

**Food processing plants.** L. J. Turney (*Industr. Engng Chem.*, 1955, **47**, 1366—1367).—The use of plastics as materials of construction in food processing plants is discussed, particularly for containers, piping, protective coating, and flooring. The advantages and limitations of plastics are outlined. O. M. Whitton.

**Humidification of cold storages: The jacket system.** C. P. Lentz (*Canad. J. Technol.*, 1955, **33**, 265—278).—By the use of the jacketed room, the surface desiccation of frozen products, e.g., fish, during storage can be reduced to negligible levels and the problem of deterioration of insulation resulting from moisture condensation can be overcome. R.H. during tests were between 89 and 98%, spatial variations in room temp. were generally less than 2°F., and velocity of air movement was negligible. Reduced frosting of the cooling coils in jacketed systems may offset most of the 10—15% increase in operating cost resulting from the increased air circulation required. The application of the test results in the design of jacketed storage rooms is discussed. O. M. Whitton.

**Air-treatment and air-conditioning installations in the food industry.** O. Kiefer (*Fette Seifen Anstrichmittel*, 1954, **56**, 1021—1023).—The applications of air-treatment in various processes of food production as well as in the storage and distribution of foodstuffs are discussed. Air-conditioning plants are described and illustrated. L. S.

**Natural food poisons, *Amanita* toxins in mushrooms.** S. S. Block, R. L. Stephens, and W. A. Murrill (*J. agric. Food Chem.*, 1955, **3**, 584—587).—A chromatographic procedure is described for the identification of toxins occurring in *Amanita phalloides*, *A. verna*, and *A. virosa* etc., 46 species of fungi, which included 13 species of Amanita, being tested. The spot colours and $R_F$ values are listed. Tested by intravenous injection into mice, the only fungi producing death were *A. verna* and *A. tenuifolia* both of which gave chromatographic tests for the amanita toxins. *A. verna* contained α- and β-amanitine, *A. tenuifolia* gave the chromatographic spot for β-amanitine only. Neither contained phalloidine. (29 references.) E. M. J.

**Potato store-houses.** N. Nicolaisen and L. Nicolaisen-Scupian (*Kältetechnik*, 1955, **7**, 299—303).—Air-conditioned or refrigerated stores have been inspected by a working party, in Holland, Denmark and Germany, where their use for food, as well as seed potatoes is extending. The best conditions are a temp. of 3—4° and R.H. ~90%. A. R. Pearson.

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**Venting material [e.g., grain] to improve and preserve its quality during storage or transport.** R. R. Anstee, C. L. Edmunds and Victor E. Smith (B.P. 730,181, 27.12.51).—A tube 3 ft. long made of Zn sheet of thickness $\frac{1}{32}$ in. tapers from 2 to 0·5 in. in diam. The smaller end is fitted with a conical cap of mild steel, and the wall is perforated with holes of diam. $\frac{1}{8}$ in. The tube is thrust into a sack of grain by a handle which can be attached to the upper end by a spigot; the latter is removed to provide a freely venting tube. J. Roberts.

**Production of ingredients for foodstuffs.** Nutrex, Ltd., P. S. Jewell and James G. T. King (inventors with N. Bezzant) (B.P. 730,182, 11.2.52).—Wheat or rye flour is treated at 28—36 (31—33)° in presence of water with the inoculation emulsion of B.P. 695,145, to give a liquid proteinaceous product containing all the proteins and the gluten, suitable for use as an ingredient (emulsifying agent) in foodstuffs (salad creams). F. R. Basford.

**[Automatic] preparation of bread dough in bakeries.** Baker Perkins, Ltd. (Inventor: J. E. Pointon) (B.P. 730,727, 27.10.52).—Dough pans are mounted on a closed endless track on the inner side of which kneading and knocking-back machines are stationed. The ingredients are charged to the pans, which pass under the kneader, then through proving stages, to the knocking-back stage and are then emptied before returning to the starting point for re-charging. K. Ridgway.

**Apparatus for boiling a sugar mass.** Hansella-Werk Albert Henkel A.-G. (B.P. 730,735, 13.3.53. Ger., 17.10.52).—An apparatus for vac.- or atm.-pressure-boiling of sugar has a boiling coil heated by steam, opening into a chamber which may be optionally connected to a vac. pump. The outlet in the base of the chamber surmounts a detachable catching vessel which is also connected to a vac. for discharging the finished product. K. Ridgway.

**Base material for thick sauces.** William Evans & Co. (Hereford and Devon), Ltd. and R. S. Potter (B.P. 732,983, 25.7.52).—A base material for thick sauces is prepared by cooking apple pomace in an aq. alkaline medium (at pH 8·5—11·0) to dissolve pectins, disintegrating the resulting pulp to form a stable suspension, and then acidifying (pH 2·5—4·5) to reprecipitate pectins in the interstices of the cellular mass. J. S. C.

**Feeding fruit [e.g., oranges] or like articles to a pulp machine.** T. White & Sons, Ltd. (Inventor: Robert S. Anderson) (B.P. 731,351, 7.12.51). K. Ridgway.

**Cutting or slicing vegetables, etc.** W. R. Gandion and M. Heaton (B.P. 730,419, 20.2.53).—A potato, to be cut into slices which have a network of holes, projects through the centre of a rotating disc, which carries a second planet disc geared to rotate at double the speed of the first, and offset from its centre. The second disc carries two cutters with corrugated edges. Each slice of potato is corrugated on its opposite sides at right angles, so that holes appear where the bases of the corrugations meet. K. Ridgway.

**Machine for sorting vegetables such as peas.** Craven & Nicholas (Engng) Ltd. (Inventors: A. J. Nicholas and J. A. Craven) (B.P. 730,434, 12.5.53).—A conventional cylindrical rotary separator, carrying internal spikes and a brush to remove any peas picked up by the spikes, suffers from the disadvantage that a large cylinder cannot be accurately made. The brush assembly is therefore mounted resiliently and carried on pivoted levers which are spring-loaded to urge the brush into contact with the cylinder, regardless of its radial position. K. RIDGWAY.

**Apparatus for detecting variation of surface characterisitcs of objects [particularly for sorting peas, beans, and other seeds].** G. M. Baigent (B.P. 730,177, 19.7.51).—The apparatus described in B.P. 712,113 is fitted with a light-interrupting device comprising a central disc with a thinner peripheral extension which is formed with a series of slots. J. M. JACOBS.

**[Control of] vegetable extracting apparatus.** G. Bredt, E. G. von Langen and O. von Loessl (trading as Pfeifer & Langen Köln, u. Braunschweigische Maschinenbauanstalt) (B.P. 731,982, 17.2.53. Ger., 28.2.52).—In a single-column sugar-beet extraction column the supply of extracting water is controlled to give a constant liquid-level in the tower, and the withdrawal of raw juice separately controlled to give a constant concentration. Efficiency of operation is increased. K. RIDGWAY.

**Stabilisation of wine and other fermented liquors.** W. & A. Gilbey, Ltd. (Inventor: W. A. Wiseman) (B.P. 732,543, 13.2.52).—Wine is stabilised by bringing into contact with sulphonated polystyrene ion-exchange resin (of B.P. 577,707) which has been regenerated with acid-free, aq. Na salt (NaCl) solution. The wine may also be conditioned (before or after treatment) by addition of $SO_2$ (50—450 p.p.m.) or (alkali metal) metabisulphite. F. R. BASFORD.

**Manufacture of beer.** Dominion Breweries, Ltd. (B.P. 730,207, 7.8.52. N.Z., 28.9.51).—Before fermentation the wort is chilled from its b.p. to a temp. just above its f.p. and is maintained there until stabilised. All material coming out of solution is filtered off and the filtrate is reheated to the fermentation temp. A more mature and stabilised beer is produced. J. ROBERTS.

**Malting and drying apparatus.** Iain M. Stewart, R. Boby, Ltd., and J. P. Wesson (B.P. 729,770, 29.9.52).—Two auxiliary perforated floors are fixed parallel to and on opposite sides of the main central floor which is mounted inside a drum. Two grain compartments are thus formed and the capacity is increased. Valved passages at one end of the drum on both sides of the main floor serve as inlets or outlets for the air only when the passage is below the floor. J. ROBERTS.

**Apparatus for oxidative fermentation of alcohols.** H. Els, trading as Heinrich Fings (B.P. 731,804, 16.8.51. Neth., 18.8.50).—In the manufacture of vinegar, the air supplied to the shavings carrying the bacteria is pretreated by passing it through a tank containing a quantity of fermenting liquor by means of a pulsating pump so that the temp. of the air is raised to fermentation temp. This prevents cooling of the bacteria on the shavings and also removes toxic substances from the air. K. RIDGWAY.

**Dry imitation vinegar.** Diamond State Products, Inc. (Inventors: T. C. Kmieciak and K. T. Farrell) (B.P. 732,837, 3.6.52).—Acetic acid (90—120) and fruit (apple) essence (15 c.c.) are adsorbed on solid binder comprising (DL-)malic acid (150—300) and sugar (300—450 g.), viz., lactose, glucose or sucrose, to provide a dry, rehydratable condiment. F. R. BASFORD.

**Device for making tea, coffee or like beverages.** J. H. Chignall and A. E. Medway & Co., Ltd. (B.P. 729,181, 4.3.53). K. RIDGWAY.

**Sterilisation of liquids [e.g., milk].** Alpura A.-G. (B.P. 731,894, 4.8.53. Switz., 22.8.52).—Milk is sterilised without alteration of flavour by passing it at 100° through a flat parallel-sided pipe section. Steam is injected through staggered rows of circular holes in the flat sides, and through longitudinal slots in the shorter sides, heating the milk to 140°. It then passes to an expansion chamber where the pressure is suddenly reduced down to atmospheric to flash off vapour and reduce the temp. rapidly. K. RIDGWAY.

**Preparation of fat-containing, stable, milk- or cream-like product from an animal milk product of low fat content.** A.-B. Separator (Inventor: A. G. Borck) (B.P. 730,887, 26.8.52).—Animal milk containing little or no fat is added at $(t + 5)°$ to $(t + 15)°$ to animal and/or vegetable oil or fat (crystallisation temp $t°$) at higher temp. and the emulsion formed by homogenisation under pressure and containing milk in the disperse phase (with oil or fat concn. 25—80%), is inverted (after adding more milk), to give a stable, milk- or cream-like product containing 5—30% of fat (in the disperse phase), suitable for use as cattle feed, for making cattle food cake, powder milk products, cheese, etc. F. R. BASFORD.

**Production of comestible materials.** Midland Counties Dairy, Ltd. (Inventor: George Edward Taylor) (B.P. 733,656, 3.12.52).—An apparatus for the extrusion of pre-determined lengths of comestible material, e.g., ice-cream, through two outlets, is described. J. S. C.

**Method and apparatus for manufacturing frozen confections.** Eskimo Pie Corp. (B.P. 731,749, 7.4.53. U.S., 14.8.52).—Feed material (e.g., ice-cream) is passed in plastic form from a continuous freezer via a discharge orifice. It is severed into portions and each portion drops on to an endless flexible metallic carrier which is maintained at a temp. of below −35°F. to ensure that the partially frozen units are sufficiently bonded to allow successive inversions. The units are released from the carrier by an electrically heated wire and fall to another conveyor for movement to an enrobing machine or packing station. Fans recirculate refrigerated air through the apparatus, which is enclosed in a chamber of brine tubes. J. ROBERTS.

**Packaging of ice-cream in frusto-conical containers.** Rose Brothers (Gainsborough), Ltd. (Inventors: A. G. Rose, J. A. Gilbert, A. S. Curtis and R. F. Gainsborough) (B.P. 731,158, 22.11.51).—A device is described for the insertion of lids into ice-cream tubs. J. ROBERTS.

**Preservation of fish.** H. R. Pauley (Inventors: Charles B. Stevenson and J. A. Hodges) (B.P. 730,217, 17.10.52).—Tuna fish is eviscerated and boned, then the skinned loins are cut into pieces, optionally dressed with brine, cooked (to coagulate proteins), washed (sprayed) with cold water, cleaned and canned. F. R. BASFORD.

**Stabilised monoester compositions.** Eastman Kodak Co., Assees. of N. H. Kuhrt (B.P. 731,177, 21.10.52. U.S., 26.10.51).—Fatty acid monoesters (**I**) (obtained by esterification of fatty acid with a polyhydric alcohol or by ester interchange), e.g., cottonseed oil or lard monoglyceride, also triglyceride compositions containing **I**, are stabilised against rancidification by compounding with glycine and $H_3PO_4$ (≯2 pt. per pt. of glycine) or salts of these. More specifically there is provided a shortening composition fortified with **I** 0·5—5% and stabilised with glycine 0·00005—0·02% and $H_3PO_4$ 0·00005—0·02 wt.%. F. R. BASFORD.

**Tempering of chocolate and similar masses.** Mikrovaerk A/S (B.P. 730,535, 25.11.53. Den., 27.11.52).—A large no. of tubes is mounted vertically between two horizontal circular tube plates, and two such assemblies are mounted in a casing. The upper one has a heating medium, e.g., water at 31°, surrounding the tubes, and the lower a cooling medium, e.g., cold water. The flow through the tubes of the chocolate mass to be tempered is made pulsatory by a rotating plate, covering the tube mouths, and having an open sector and a scraper. The scraper passes the product over a temp.-sensitive element which controls the temp. of the heating and/or cooling media. K. RIDGWAY.

# 3.—SANITATION

**Industrial hygiene.** H. H. [Schrenk (*Industr. Engng Chem.*, 1955, **47**, No. 10, 95A—96A).—Basic considerations in evaluating health hazards of food packaging materials and insect repellants are presented. O. M. WHITTON.

**Mechanism of insecticide action. Preparation, purification, isomerisation and biological properties of octamethylpyrophosphoramide *N*-oxide.** Hiroshi Tsuyuki, M. A. Stahmann and J. E. Casida (*J. agric. Food Chem.*, 1955, **3**, 922—932).—As a metabolic activation of octamethylpyrophosphoramide (schradan) is required before it is toxic to insects and as the product formed is identical with chemically oxidised (neutral $KMnO_4$) derivatives, such derivatives were tested as contact and systemic insecticides. The specificity of the permanganate oxidation products for choline-esterase inhibition indicated that the oxidation of only one N yields the most effective anticholine-esterase whereas further oxidation yields other derivatives which liberate formaldehyde on acid hydrolysis, but are of lower anticholine-esterase activity. Oxidation of schradan with other oxidising agents, e.g., hypochlorite, peracetic acid under anhydrous conditions, etc., results in re-arrangement of the octamethylpyrophosphoramide *N*-oxide mol. Possible structures of compounds are presented and discussed. Chromatographic purification of oxidation products yielded an unstable, active anticholine-esterase and a more stable oxidation product. (37 references.) E. M. J.

**Problems connected with ethylene dibromide fumigation of cereals. I. Sorption of ethylene dibromide by grain.** E. Olomucki and A. Bondi. **II. Feeding experiments with laying hens.** A. Bondi, E. Olomucki and M. Calderon (*J. Sci. Food Agric.*, 1955, **6**, 592—600,

600—602).—I. Most of the Br in $C_2H_4Br_2$-fumigated grain is unchanged $C_2H_4Br_2$; a small amount of residual non-volatile bromide, mainly resulting from reaction of $C_2H_4Br_2$ with the protein fraction, was also found. Differences in sorption were correlated with varying morphological structures of grains, in particular differential seed-coat permeability. Airing of fumigated grain for 6—7 weeks results in loss of the volatile Br component.

II. $C_2H_4Br_2$ absorbed by grain is injurious to egg production. Hens fed grain containing 200 p.p.m. of free $C_2H_4Br_2$ for 56 days (or 300 p.p.m. for 46 days) stopped laying completely. After 12 weeks, feeding of grain containing 10 p.p.m. of $C_2H_4Br_2$ causes a slight diminution of egg size. The residual (non-volatile) bromide appears to have no injurious effect. J. S. C.

**Control of insect pests in grains stored in insecticide-impregnated jute bags.** M. Muthu and S. V. Pingale (*J. Sci. Food Agric.*, 1955, **6**, 637—640).—Five chemicals ($CH_3Br$, a 3 : 1 mixture of $C_2H_4Cl_2$ and $CCl_4$, $CCl_4$, acrylonitrile, and $C_2H_4Br_2$) were injected into grain stored in insecticide-impregnated bags in non-airtight conditions. It was found that complete control of insect pests studied [*Calandra* (*Sitophilus*) *oryzae*, (L.), *Rhizopertha dominica*, (F.), and *Tribolium castaneum*, (Hub.)] was obtained by injection of 10 c.c. of $C_2H_4Br_2$ per 200-lb. bag. For bags arranged in stacks, a dose of 7·5 c.c. ensured complete insect mortality. The quantities of chemical absorbed by the grain were measured and found to be within the permissible limits. J. S. C.

**Protection of stored grain with sprays of pyrethrins–piperonyl butoxide emulsion.** W. E. Dove and H. O. Schroeder (*J. agric. Food Chem.*, 1955, **3**, 932—936).—Laboratory tests made with sprays containing pyrethrum and piperonyl butoxide applied directly to grain indicated that water emulsions were more staisfactory and more lasting in their effect to control insects than sprays containing petroleum oil or vegetable oil as diluents. An oil-free emulsion containing 2% of piperonyl butoxide and 0·2% of pyrethrum applied at the rate of 5 gal. per 1000 bushels of grain was adequate for the protection of stored grain. E. M. J.

**Absorption and metabolism of [$^{14}$C]-pyrethroids by adult house fly, *Musca domestica*, L., *in vivo*.** F. P. W. Winteringham, A. Harrison and P. M. Bridges (*Biochem. J.*, 1955, **61**, 359—367).—A natural mixture of pyrethroids, "labelled" biosynthetically with $^{14}$C is resolved by reversed-phase paper chromatography into chrysanthemic esters, pyrethric esters, and unidentified non-insecticidal impurities. After topical application of these esters or of allethrin "labelled" with $^{14}$C in the alcoholic component of the mol. to adult house flies (or after injection into the insects), significant and comparable fractions of all of them are metabolised to relatively non-insecticidal products within 24 hr. When the synergist, piperonyl cyclonene, is applied simultaneously with the pyrethroids, the metabolism is markedly inhibited, but least effectively with allethrin. Synergism may involve interference with the natural detoxication mechanism of the house fly. Absorption of the topically-applied pyrethroids is almost complete in 24 hr., and is non-selective from a mixture of esters. Addition of piperonyl cyclonene always retards absorption, probably by dilution of the pyrethroid on the insect surface. J. N. Ashley.

**Comparison of ethylenedioxyphenyl and methylenedioxyphenyl compounds as extenders for pyrethrins.** E. A. Prill and William Richard Smith (*Contr. Boyce Thompson Inst.*, 1955, **18**, 187—192).—Using, as reference compounds, *N-iso*-butyl-3 : 4-methylenedioxycinnamide (fagaramide) and 2-(3 : 4-methylenedioxyphenyl)-5 : 5-diethyl-*m*-dioxan, the corresponding ethylenedioxyphenyl (**I**) compounds, viz., *N-iso*butyl-1 : 4-benzdioxan-6-acrylamide and 2-(1 : 4-benzdioxan-6-yl)-5 : 5-diethyl-*m*-dioxan, were prepared and tested for synergistic activity with pyrethrins against house flies (*Musca domestica*, L.). They were found to be inactive in this respect and it is concluded that synergetic activity is probably dependent on some characteristic of the methylenedioxyphenyl nucleus as such. (13 references.) J. S. C.

**Chromatographic [2 : 4-dinitrophenylhydrazone] method for determination of allethrin.** N. Green and M. S. Schechter (*Analyt. Chem.*, 1955, **27**, 1261—1265).—The method is based on the conversion of allethrin to the 2 : 4-dinitrophenylhydrazone deriv. (**I**), which is separated on a silicic acid column and determined by gravimetric or colorimetric means. The **I** band on the column is well separated from interfering material, and is easily followed visually. Results from technical allethrin samples, using the gravimetric conclusion, are in close agreement with the ethylenediamine method, and the colorimetric procedure has an accuracy of approx. ±10%. G. P. Cook.

**Infra-red spectrophotometric determination of allethrin.** S. K. Freeman (*Analyt. Chem.*, 1955, **27**, 1268—1274).—Allethrin is determined by measurement of the 5·81 $\mu$ band, a wavelength at which only allethrolone, of the four possible impurities in the insecticide, interferes. The i.r. absorption spectra of these impurities, allethrolone, and chrysanthemum-monocarboxylic acid and its acid chloride and anhydride were studied, and techniques for their determination, as well as means to differentiate between *cis*- and *trans*-allethrin are described. More than fifty commercial samples were analysed, and, of twelve chosen as representative, results were in close agreement with the ethylenediamine chemical method, with one exception (difference of 0·8% on assay). The commercial samples displayed a fairly constant (20 : 80) *cis–trans* ratio, and the anhydride was a minor constituent of these samples. G. P. Cook.

**Resistance of insects to insecticides.** J. Lhoste (*Chim. et Industr.*, 1955, **74**, 681—692).—The literature dealing with various aspects (morphological, chemical, physiological) of the resistance of insects to insecticides, the effects of different insecticides, etc., are reviewed. (78 references.) J. M. Jacobs.

**Fly repellents.** D. E. Howell and L. D. Goodhue (*Soap, N.Y.*, 1955, **31**, No. 10, 181, 185, 187, 189, 221).—Results of laboratory and field tests on the efficiency as fly repellents of di-*n*-propyl *iso*-cinchomerate (**I**), 2 : 3 : 4 : 5-bis(but-2-enylene)tetrahydrofurfural (**II**) and 2 : 3 : 4 : 5-bis(but-2-enylene)tetrahydrofurfuryl alcohol (**III**) are reported. They provide good protection under most conditions. In outdoor conditions, the duration of repellency can be greatly improved by addition of stabilisers. Wettable powder formulations are superior to other types of sprays. **I** is particularly repellent to house flies, while **II** and **III** are much more repellent to stable flies. Combinations of **I** with either **II** or **III** show strong synergistic action. The pyrethrum synergist, *N*-octyl-*bicyclo*-heptene dicarboximide, increases the effectiveness of all these repellents. J. S. C.

**Toxicity of trinitrobenzene–aniline complex, a rodent repellent.** R. W. Fogleman, J. R. Elsea, O. E. Paynter and W. Kundzins (*J. agric. Food Chem.*, 1955, **3**, 936—939).—In rats the acute oral $LD_{50}$ for trinitrobenzene–aniline complex (TNBAC) was 375 mg. per kg. of body wt.; for trinitrobenzene (TNB) 505 mg. per kg. In dogs, repeated daily, oral doses >25 mg. per kg. of TNBAC produced signs of gros toxicity and hæmatological changes. Using each of these formulations for dermal application data indicated that TNBAC was absorbed and produced systemic effects, but no skin irritation. Repeated doses are cumulative. TNBAC should not be used in granaries or barns where animal or human food is stored; care should be taken by individuals to prevent exposure to the repellent, and contamination of clothing. E. M. J.

**Identification of rodent fur hairs.** Dorothy B. Scott (*J. Ass. off. agric. Chem. Wash.*, 1955, **38**, 503—506).—The fur hairs of the rat, mouse, rabbit, squirrel and musk-rat were mounted in glycerol jelly and examined microscopically without further treatment. The structure of the medullary unit is of diagnostic value. Photographs (except for mouse hair, which is similar to rat fur hair) are reproduced. A. A. Eldridge.

**Suggested procedure for evaluation of biological oxidation of organic chemicals.** E. J. Mills, jun., and V. T. Stack, jun. (*Sewage industr. Wastes*, 1955, **27**, 1061—1064).—It was found that micro-organisms employed as seed can constitute a variable in B.O.D. determination with certain org. chemicals as a result of acclimatisation. To overcome this possible source of error, a procedure is recommended whereby a long-term B.O.D. curve is determined using as seed an unacclimatised culture. If this fails to give satisfactory results, a procedure is defined in detail for acclimatising organisms from river water, and the B.O.D. determined using the acclimatised organisms as seed. If acclimatisation is not obtained within 100 days, it is assumed that the chemical tested cannot be oxidised by aërobic micro-organisms. J. S. C.

**Determination of biochemical oxygen demand by a re-aëration technique.** H. L. Elmore (*Sewage industr. Wastes*, 1955, **27**, 993—1002).—The technique described is primarily designed to determine the B.O.D. of samples derived from streams and reservoirs, and consists of incubating several sealed bottles and a larger, unsealed bottle, of a sample. The dissolved $O_2$ is measured by the Winkler method initially and at various intervals. When total depletion of the $O_2$ is approached, another set of bottles is prepared from the unsealed bottle and the procedure continued through any time interval desired. The re-aëration procedure applied to the samples consists of removal of supersaturated $O_2$ by vac. (temp. of sample 23—25°), and aëration for 15 min. with a diffusion stone. A device comprising a magnetic stirrer, a thermostat heater and the necessary connexions for air and water flow is described for carrying out the procedures. (16 references.) J. S. C.

**Research on activated sludge. V. Rate of biochemical oxidation.** A. Pasveer (*Sewage industr. Wastes*, 1955, **27**, 783—792; cf. J.S.F.A. Abstr., 1954, ii, 60).—A series of laboratory tests, in which the B.O.D. load and oxygenation capacity of activated sludge treatment

were varied, showed that the rate of biochemical oxidation can be increased up to at least 50 times the rate at which the process goes on in a normally-loaded activated sludge tank in practice. The limiting factor in the process is oxygen supply. Only in the smallest flocs and with strong turbulence is the bulk of the floc active in biochemical oxidation. Because of this, the variation of biological oxidation in respect of temp. is comparatively small. J. S. C.

**Studies of biochemical oxidation by direct methods. V. Effect of various seed materials on rates of oxidation of industrial wastes and organic compounds.** I. Gellman and H. Heukelekian (*Sewage industr. Wastes*, 1955, **27**, 793—801).—A comparative study was made of the effect of a no. of seed materials on the oxidation of industrial wastes (e.g., sulphite pulp cooking liquors from paper mills, spent yeast, nutrient waste, etc.) and org. compounds (including Me, Et, Pr$^i$ and Bu alcohols. The (decreasing) order of effectiveness of the seeds was: acclimatised activated sludge, mixed dispersed growth, settled dispersed growth, normal activated sludge, and normal sewage seed. Differences in composition of the waste materials and org. compounds studied were reflected in differences of oxidisability; in particular, it was found that the 1- and 3-C alcohols were less readily oxidised than the 2- and 4-C alcohols, with a corresponding decrease in the rate of sludge accumulation, indicating a slowing down of cell formation. J. S. C.

**Application of biological treatment to industrial wastes. I. Kinetics and equilibria of oxidative treatment.** R. F. Weston and W. W. Eckenfelder (*Sewage industr. Wastes*, 1955, **27**, 802—819).—A theoretical account, based on available published data, of the oxidative biological waste treatment indicates that it proceeds in three phases: (*a*) initial rapid removal of B.O.D., on contacting biologically active sludge, due to reaction between the enzymes of the sludge and the org. constituents of the waste, (*b*) removal of B.O.D. in direct proportion to biological cell growth, and (*c*) oxidation of biological cell material with concurrent slow removal of B.O.D. (26 references.) J. S. C.

**Citrus waste water treatment of activated sludge.** M. H. Dougherty, R. W. Wolford and R. R. McNary (*Sewage industr. Wastes*, 1955, **27**, 821—826).—Laboratory studies indicate the feasibility of treating waste water from citrus processing by the activated sludge process. It was found unnecessary to add nitrogenous compounds to supplement the low N-compound content of the waste, but the addition of small amounts of alkali may be necessary to adjust the pH value to above 7·0. The excess sludge produced may be dried and used in animal feeds, being a source of B-vitamins. J. S. C.

**Cannery waste disposal by irrigation.** G. H. Dunstan and J. V. Lunsford (*Sewage industr. Wastes*, 1955, **27**, 827—834).—Experience in the U.S.A. on the disposal of cannery effluents by irrigation is reviewed and an experimental irrigation of test plots by a pea-canning factory is described. Of two identical soils, that covered with vegetation proved to utilise the most effluent, and permanent pasture grasses were able to absorb a heavier org. loading than lucerne under identical conditions. (8 references.) J. S. C.

**Organic nitrogen preferences of *Zoogloea ramigera*.** L. G. Rich (*Appl. Microbiol.*, 1955, **3**, 20—25).—Cultures of the organism obtained from sewage trickling filters were grown on media containing various N sources and respiration rates were examined by periodic polarographic determinations of the [$O_2$] in the substrate. *Z. ramigera* utilised proteoses and peptones more readily than purified proteins or individual amino-acids. Aërobic respiration proceeded on purified protein media, rates of respiration varying widely with the nature of the protein. In concn. >25 p.p.m. in the substrate $NH_3$ was toxic to the organism. A. G. POLLARD.

**Two-stage operation of activated sludge plants.** K. Imhoff (*Sewage industr. Wastes*, 1955, **27**, 431—433).—A critical review of two-stage operation of activated sludge plants. The two-stage plant differs from the normal excess sludge method of operation in that in each stage there is an aëration tank and secondary settling tank. The advantage of the process is that the excess sludge for disposal contains less water. A smaller aëration unit can be used by a suitable by-pass and any degree of partial biological purification can be obtained. J.A.C. ABSTR.

**Importance of air disinfection in medical science.** H. C. Bartelma (*Pharm. Weekbl.*, 1955, **90**, 677—689).—A review covering general considerations, methods for bacteriological analysis of air, including a description of the Bourdillon "slit sampler" and its use, and the use of dust-laying oils and aërosol sprays. (28 references.) P. S. ARUP.

**[Preparation of a] phosphonic ester.** Farbenfabriken Bayer A.-G. (B.P. 731,506, 9.9.53. Ger., 20.9.52).—The prep. of *Me*$_2$ *(2 : 2 : 2-trichloro-1-hydroxyethyl)phosphonate* (**I**) and its use as an insecticide (against flies) are claimed. Thus, chloral (75) is added slowly to dimethyl phosphite (60) at initially 25°, temp. being allowed to rise to ⩾50—60°, then a solution of the resulting oil in benzene is washed with aq. $NaHCO_3$, dried, concentrated, and the concentrate cooled, to give **I** (90 g.), m.p. 81°. A 1% solution of **I** in $(CH_2Cl)_2$ sprayed at eventide on the walls of a room inhabited by flies results in their complete elimination by morning. F. R. BASFORD.

**Production of antibiotic agent.** Schenley Industries, Inc. (B.P. 722,433, 1.2.52. U.S., 16.10.51).—*Fluvomycin*, $C_{13-14}H_{18-20}O_{3-5}N_2$, mol. wt. 261—272, C 50—52, H 6·8—7, N 7·9—8·5%, $[\alpha]_D^{20}$ + 78°, active against Gram-positive and Gram-negative micro-organisms, also to pathogenic fungi (including *Candida albicans*), is obtained by propagating *Bacillus subtilis* in nutrient medium at pH 5—9 under aerobic conditions, then adsorbing the product on C, and eluting therefrom with aq. acetone or aq. alcohol. J.A.C. ABSTR.

## 4.—APPARATUS AND UNCLASSIFIED

**Estimation of total nitrogen in biological material.** E. Zymny (*Dtsch. ApothZtg*, 1955, **95**, 1035—1036).—A brief review of the disadvantages of the Kjeldahl method for the estimation of total N is given, and an alternative Autenrieth–Taege procedure (*Münch. med. Wschr.*, 1922, No. 31, 1141—1143), using conc. $H_2SO_4$ and a Se catalyst, is also described. G. R. WHALLEY.

**Paper chromatography of uronic acids.** R. A. Edington and E. Percival (*J. chem. Soc.*, 1955, 3554—3555).—$R_G$ values are given for eleven methylated uronic acids and lactones, using aged or otherwise equilibrated solvent mixtures [4 : 1 : 5 BuOH–AcOH–water (**I**) and 100 : 23 : 77 BuOH–formic acid–water (**II**)]. **II** gives more consistent results (±3%), is faster, gives better separation, and is less liable to cause badly-shaped spots. Aq. aniline oxalate and *p*-anisidine hydrochloride–butanol are used as spray reagents. M. DAVIS.

**Dependence of $R_F$ values of amino-acids on the shape and size of the filter paper.** H. C. Chakrabortty and D. P. Burma (*J. Indian chem. Soc.*, 1955, **32**, 533—536).—The $R_F$ values of serine and valine are affected very little by the strip width or the sector angle of the filter paper. The values on strips are equal to the squares of those on sectors. O. M. WHITTON.

**Colorimetric phosphorus determination in phospholipins.** A. E. F. H. Meijer (*Proc. K. Ned. Akad. Wet.*, 1955, **B, 58**, 272—281).—A procedure is outlined for the colorimetric determination of P in phospholipins. Quinol, aminonaphtholsulphonic acid and $SnCl_2$ are investigated as reducing agents. In the molybdenum blue procedure, after digestion with $H_2SO_4$ and $H_2O_2$, the solution is reduced by quinol, followed by determination of the extinction coeff. and comparison with standards. Temp. and pH are important. $SnCl_2$ is too powerful as a reducing agent, but aminonaphtholsulphonic acid gives results equivalent to those with quinol. The average relative error is <5%, and 0·1—0·8 mg. of P can be determined. (23 references.) R. J. MAGEE.

**Spectrophotometric estimation of acid and alkaline phosphatases with *o*-carboxyphenyl phosphate.** H. Brandenberger and W. H. Weihe (*Helv. chim. Acta*, 1955, **38**, 1347—1351).—Comparison is made of the rate of enzymic hydrolysis of *o*-carboxylphenyl phosphate and other substrates used for phosphatase determinations. The results show that an acid phosphatase, obtained from wheat germ, caused a more rapid hydrolysis than that obtained with β-glycerophosphate, phenyl phosphate and phenolphthalein diphosphate, and this decomposition was slightly slower with β-glycerophosphate when treated with two alkaline phosphatases, although this was still more than ten times as rapid as with phenolphthalein diphosphate. *o*-Carboxyphenyl phosphate, however, is still preferred, in view of the ease of determination, which requires absorption measurement at 298 mμ. with a Beckman spectrophotometer, using 2 ml. of an acetate buffer solution of pH 5, 0·5 ml. of the required substrate solution and 0·5 ml. of the enzyme solution. G. R. WHALLEY.

**Quantitative determination of water.** A.-B. Pharmacia, Assees. of E. Blomgren and H. Jenner (B.P. 722,983, 9.12.52. Sw., 10.2.51).—A Karl Fischer reagent (**I**), e.g., a solution of I, $SO_2$ and pyridine in a hydroxylated solvent, e.g., methanol, for use in the quant. determination of water, is stabilised against decrease in titre during storage, due to side-reactions, by addition of a stabiliser identical with a reaction product of the side-reaction, e.g., pyridine hydriodide (**II**) or water. Thus, the titre of **I** prepared from I 84, $SO_2$ 85, pyridine 308, methanol (690 c.c.), and **II** 70 g. remains constant with time. J.A.C. ABSTR.

# ıETY OF CHEMICAL INDUSTRY

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*Title.*—This should be concise and explanatory of the purpose of the paper. Where a related series of papers is submitted each individual paper should have the same general heading, followed by a series number and title of the part.

*Synopsis.*—A short synopsis of the work, drawing attention to salient points, and intelligible without reference to the paper itself, should be given separately at the beginning of the paper.

*Introduction.*—The aim of the investigation should be given and also a brief statement of previous relevant work with references.

*Experimental.*—The methods and materials used should be clearly stated in sufficient detail to permit the work to be repeated if desired. Only new techniques need to be described in detail, but known methods should have adequate references.

*Results.*—These should be presented concisely, using tables or illustrations for clarity. Adequate indication of the level of experimental error and the statistical significance of results should be given. Only in exceptional cases will tables *and* graphs derived from them be accepted for publication.

*Discussion.*—In general, the discussion and interpretation of results should follow their presentation, in a separate section.

*Conclusions.*

*Acknowledgments.*

*References.*

### IV. Typescript

(*a*) All manuscripts should be typed in double spacing on one side of the paper only and adequate margins should be left. One copy should be retained by the author, and the top copy should be sent to the appropriate Editor.

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