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EFFECTS OF SALT AND OTHER FERTILISERS ON YIELD AND MINERAL COMPOSITION OF FORAGE CROPS. I.—Turnips

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The literature concerning the influence of sodium on phosphorus utilisation by plants has been reviewed together with the frequent suggestions that sodium may be able partially to replace potassium in the nutrition of Brassica crops. It has been found that salt increased the yield of turnips in each of four experiments by greater amounts than did muriate of potash. There were consistent negative interactions between salt and potassium. Muriate of potash greatly depressed sodium uptake and salt increased it by about 50%. Salt was shown to improve phosphorus uptake and there were negative interactions of importance between salt and superphosphate for both the yield and phosphorus content of the roots.

Introduction

It is now generally accepted that sodium is an important nutrient for sugar beet and other members of the Chenopodiacea. Suggestions have frequently been made that sodium may be of value for many other plants and in particular, that a variety of Brassica crops may give responses under suitable circumstances. Thus, Harmer & Benne[1] report that when potassium is deficient there may be slight to medium responses to salt with broccoli and Brussels sprouts and that cabbage, kale, mustard and turnips respond even when there is an adequate potassium supply. Lehr,[2] Truog *et al.*,[3] Kennedy *et al.*,[4] and Larson & Pierre[5] have also published lists of crops which respond to salt and, in general, draw the same conclusions. Pizer[6] has shown that kale, swedes, rape, mustard and turnips are amongst the most tolerant of crops to the adverse effects of sea water flooding. Harmer & Benne[1] have reported large increases in the yield of cabbage from salt.

The main emphasis of most of past work with sodium has been to investigate its importance as a partial substitute for potassium. There are, however, many suggestions that applications of salt may encourage more efficient utilisation of phosphorus. As long ago as 1906, Wheeler & Hartwell[7] reported that sodium salts increased the phosphorus content of a number of crops. Collings[8] has recently concluded that sodium is valuable ' to maintain a high degree of availability of phosphorus . . . and may increase the availability of phosphorus that is tied up in insoluble form '.

Hebert[9] found that sodium nitrate extracted three times as much phosphorus from soils as did calcium nitrate. Using the same salts at concentrations equivalent to the nitrate level in 22 Dutch soils, Lehr & Wesemael[10] reported that sodium nitrate was superior to the extent of 80–90% and that sodium salts leached through the soils maintained a greater and more prolonged release of phosphorus compared with calcium salts. In subsequent Neubauer tests they found[11] that phosphorus solubility was 64% greater and phosphorus uptake 45% higher if sodium nitrate replaced calcium nitrate. Tobia & Milad[12] have found that sodium and potassium carbonates applied to Egyptian alkali soils increased the concentration of water-soluble phosphorus, whereas calcium and magnesium salts depressed it.

Kibe *et al.*[13] showed that the available phosphorus in pot experiments with wheat was increased by an application of 0·2% sodium chloride. On the other hand, Scharrer & Schreiber[14] had found that sodium chloride reduced the phosphorus uptake of rye seedlings and McEvoy[15] showed that it depressed the phosphorus content of tobacco. Marshall & Sturgis[16] indicate that, when sodium nitrate is the best nitrogenous fertiliser for cotton, the soils are frequently low in phosphorus. Nicholson & Hooper[17] have suggested that the superiority of sodium and potassium nitrates over other nitrogenous fertilisers for cabbage may be due to their effect on soil phosphorus.

Way & Nelson[18] inhibited the formation of citrate-insoluble phosphorus in NP and NPK fertilisers by the addition of 1% sodium chloride to the phosphate rock prior to acidulation.

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Hamamato & Kawasaki[19] by treatment of rock phosphate with sodium chloride and steam produced a fertiliser as efficient for rice as was superphosphate. Andrews[20] has suggested that sodium additions to soil may lead to the formation of sodium fluoride which is leached out, thus reducing the creation of calcium fluoride-containing apatites of low availability.

The recent observations of Boyd *et al.*[21] who demonstrated the presence of substantial negative interactions between sodium and phosphorus, and sodium, potassium and phosphorus in a comprehensive series of sugar beet experiments which were ' too frequent to be ignored ' give added practical support to the supposition that sodium may increase the efficiency of phosphorus utilisation.

There have been no recent experiments in this country regarding the use of salt as a fertiliser for crops other than sugar and fodder beets and mangolds in recent years. This present work is concerned with its effects in association with other fertilisers on the yield and mineral composition of turnips, kale and grassland.

In 1914, Bolin[22] reported the results of ten trials in Sweden where 9 cwt. of salt gave a mean increment of 5·5 tons of swedes. Harmer & Benne[23] obtained an increase in yield of 3 tons of turnips from 5 cwt. of salt on a potassium-deficient soil. Crowther & Benzian[24] have summarised the results of a number of pre-1939 experiments where ammonium sulphate and sodium nitrate were compared as nitrogen sources and concluded that the latter was 20–25% superior for turnips. Lehr & Bussink[25] have compared sodium nitrate favourably with calcium nitrate, but Dorphpetersen & Steenbjerg[26] found negligible differences between them in many experiments except for one on a potassium-deficient soil. Holt & Volk[27] found in pot experiments that 30% of a full crop of turnips could be obtained in the complete absence of potassium.

Experimental

Two field experiments were completed in 1955 and a further two in 1956. In 1955 the layouts were in $3 \times 3 \times 3$ designs of 27 plots to investigate all combinations of 0, 3 and 6 cwt. of salt, 0, 3 and 6 cwt. of superphosphate, and 0, 1 and 2 cwt. of muriate of potash. In addition, each plot had a basal dressing of 2 cwt. of ammonium sulphate. In 1956 the design was changed to a $2 \times 2 \times 2 \times 2$ layout in four blocks of eight plots with the NPK/salt interaction confounded between each of the two replicates at each site. The treatments were the presence and absence of 4·5 cwt. of salt, 4·5 cwt. of superphosphate, 1·5 cwt. of muriate of potash and 2 cwt. of ammonium sulphate.

The crops were grown on slightly raised ridges, 27 in. apart. The fertilisers were applied by hand before the final seed-bed preparations but after the first harrowing. Singling, weeding, etc. were carried out in conjunction with the remainder of the field. There were no adverse effects on germination. The plot size was 0·01 acre, each plot being six rows wide. The centre four rows were harvested in November. The roots were sampled by taking a diagonal core from about 40 roots. Fifteen tops were selected at random from each plot for analysis.

Methods of analysis

Sodium and potassium were determined by means of the EEL flame photometer. Calcium was also estimated in a similar manner after removal of phosphorus (Hemingway[28]). The Titan yellow method of Cornfield & Pollard[29] was adapted for the determination of magnesium and the normal molybdenum blue method was used for phosphorus. One-g. samples were ashed at 500°, 1 ml. of conc. HCl was added and evaporated to dryness on a hot plate. The resulting ash was extracted with hot water and made up to 100 ml.

The scheme of analysis outlined in Table I was followed. In addition to the speed of the individual methods themselves, it has the advantage that by use of a graduated pipette, all the analyses may be commenced in one operation. One person is able to complete the entire analysis of up to 20 samples per day.

The accuracy of this scheme of analysis was assessed by the replicated analysis over a period of samples of hay and kale leaf (Table II). These errors were considered satisfactorily small in view of the speed and simplicity of the method.

Table I

Rapid scheme of analysis for sodium, potassium, calcium, magnesium and phosphorus in plant material

(1·0-g. sample ashed, to give 100 ml. solution)

	Pipetted vol., ml.	Element	Final vol., ml.	Standard curve	% Element in dry matter
1	2·5	Calcium	10	0–75 p.p.m. Ca	0– 3·0
2	2·5	Magnesium	25	0–0·1 mg. Mg	0– 0·4
3	1·0	Phosphorus	100	0–0·05 mg. P	0– 0·5
4	1·0	Sodium	10	0–5 p.p.m. Na	0– 0·5
5	1·0	Potassium	100*	0–10 p.p.m. K	0–10·0

* This solution may alternatively be obtained by diluting 1 ml. of the solution prepared for the sodium analysis with 9 ml. of water. It may also be used in those cases where the Na % exceeds 0·5%.

Table II

Replicate analyses of samples of hay and kale leaf

	Hay					Kale leaf				
	Na	K	Ca	Mg	P	Na	K	Ca	Mg	P
	(% of dry matter)					(% of dry matter)				
Mean of 10 determinations	0·186	2·38	0·31	0·169	0·308	0·234	2·09	2·31	0·110	0·269
Standard deviation as % of mean	3·32	2·47	2·21	3·83	1·94	3·31	3·53	4·59	7·16	2·44
Mean of two determinations by 'standard' methods of analysis	0·182	2·32	0·32	0·174	0·320	0·240	2·07	2·33	0·106	0·275

Results

In order to give comparable results for the two sets of experiments, the responses to 3 and 6 cwt. of salt and superphosphate and to 1 and 2 cwt. of muriate of potash in Expts. 1 and 2 have been averaged. Only consistent or important interactions have been tabulated, the remainder being very small and irregular.

Yields (Table III)

Salt has increased the yield of roots in all four experiments by amounts ranging up to 2 tons. These are to be compared with the much smaller returns from muriate of potash. There were no consistent effects on the yield of tops. Dry matter yields were affected in a similar manner. The responses to potassium and salt were not related to the 1% citric-soluble soil potassium, but none of the soils could be described as really deficient. There were large and consistent negative interactions between salt and potash and between salt and superphosphate.

Superphosphate increased yields in all but the first experiment by large and significant amounts. The responses were broadly correlated with the soil analyses, soils 2 and 4 being acutely deficient.

Plant composition

Sodium (Table IV).—Fertilisers have markedly influenced sodium uptake. In every experiment salt has increased the sodium content by large and very significant amounts, generally of the order of 50% of the mean. Conversely, muriate of potash has reduced the sodium levels, also significantly, and by almost as much. Superphosphate increased sodium uptake in all experiments except the first, which was the only one where it did not increase yields. Ammonium sulphate had only small effects. There were no important interactions.

Potassium (Table V).—Salt has had very little influence on potassium uptake, the tendency being to increase it by small amounts. The increased absorption of sodium does not therefore enable the plant to reduce its potassium requirement. Superphosphate and ammonium sulphate also had little effect. Muriate of potash did however consistently increase the potassium content of both roots and tops. The increases ranged up to 0·5% of K and were frequently significant. There were no marked interactions.

Calcium (Table VI).—The fertiliser treatments have had very little effect on the calcium contents of both roots and tops. Salt and muriate of potash tended to depress the levels.

Table III

Yields of roots and tops (tons) and soil analyses

	Roots				Tops			
Experiment	1	2	3	4	1	2	3	4
Mean yield	18·36	17·76	16·06	21·47	3·73	2·96	3·39	5·08
Response to								
Salt 4·5 cwt.	1·94	1·73	0·73	1·87*	0·25	0·01	0·11	−0·34
Muriate of potash 1·5 cwt.	0·72	0·24	1·17*	0·90	−0·59	−0·55	0·42**	−0·10
Superphosphate 4·5 cwt.	−0·74	6·39**	1·37*	5·02**	−0·45	0·56	0·22*	0·97**
Sulphate of ammonia 2·0 cwt.	—	—	−0·28	0·58	—	—	0·22*	0·14
Standard error ±	1·11	1·17	0·46	0·63	0·29	0·31	0·08	0·24
Interactions								
K/salt	−0·99	−1·92	−0·01	−1·63*	0·18	0·00	0·03	−0·53*
P/salt	−0·69	−1·65	−0·57	−2·61**	−0·50	−0·06	0·09	−1·62**
Standard error ±	1·93	2·03	0·65	0·89	0·50	0·54	0·11	0·34
Soil analysis 1% citric soluble								
P, mg.-%	13·0	4·0	9·5	3·5				
K, mg.-%	13·5	11·0	12·5	8·5				
pH	6·7	6·1	6·7	5·4				

Table IV

% Na in dry matter

	Roots				Tops			
Experiment	1	2	3	4	1	2	3	4
Mean Na %	0·120	0·146	0·175	0·134	0·171	0·332	0·226	0·402
Response to								
Salt 4·5 cwt.	0·053**	0·080**	0·044**	0·064**	0·076**	0·197**	0·098**	0·237**
Muriate of potash 1·5 cwt.	−0·049**	−0·028	−0·035**	−0·033**	−0·048**	−0·052	−0·074*	−0·078*
Superphosphate 4·5 cwt.	−0·028*	0·009	0·023	0·020	−0·039*	0·002	0·050*	0·042
Sulphate of ammonia 2·0 cwt.	—	—	0·018	−0·015	—	—	0·025	0·011
Standard error ±	0·008	0·010	0·011	0·011	0·010	0·020	0·022	0·028

Table V

% K in dry matter

	Roots				Tops			
Experiment	1	2	3	4	1	2	3	4
Mean K %	3·06	2·60	2·05	1·68	3·17	3·36	2·41	1·98
Response to								
Salt 4·5 cwt.	0·04	0·12	−0·08	0·13*	0·13	0·15	−0·03	−0·03
Muriate of potash 1·5 cwt.	0·17	0·05	0·24*	0·31**	0·48*	0·27	0·18	0·49**
Superphosphate 4·5 cwt.	−0·09	0·18	−0·05	−0·07	0·31	0·17	−0·23*	−0·19
Sulphate of ammonia 2·0 cwt.	—	—	−0·01	0·00	—	—	−0·01	−0·12
Standard error ±	0·08	0·12	0·08	0·05	0·17	0·14	0·10	0·09

Magnesium (Table VII).—Fertilisers generally reduced the magnesium content of turnips. In the case of salt the depressions were less than 0·007% of Mg. They reached 0·021% in the leaves as a result of potassium application. These effects were additive in that the potassium–salt interactions were positive, but the errors were large in proportion.

Phosphorus (Table VIII).—Salt has increased the uptake of phosphorus in each experiment. In Expts. 2 and 4 where the largest yield responses to superphosphate were found, the increases

Table VI

% Ca in dry matter

	Roots				Tops			
Experiment	1	2	3	4	1	2	3	4
Mean Ca %	0·66	0·40	0·38	0·36	2·31	2·02	1·80	1·71
Response to								
Salt 4·5 cwt.	−0·01	0·05	−0·02	−0·02	−0·14	−0·10	0·03	−0·06
Muriate of potash 1·5 cwt.	−0·01	0·01	−0·03*	−0·01	−0·08	−0·14	−0·05	−0·02
Superphosphate 4·5 cwt.	0·00	0·05	0·01	0·00	−0·10	0·02	−0·06	−0·11
Sulphate of ammonia 2·0 cwt.	—	—	−0·01	−0·03	—	—	0·15*	−0·08
Standard error ±	0·03	0·02	0·01	0·01	0·14	0·04	0·05	0·10

Table VII

% Mg in the dry matter

	Roots				Tops			
Experiment	1	2	3	4	1	2	3	4
Mean Mg %	0·103	0·082	0·081	0·075	0·204	0·208	0·132	0·135
Response to								
Salt 4·5 cwt.	−0·004	−0·003	−0·006*	−0·004	−0·007	−0·008	0·000	0·003
Muriate of potash 1·5 cwt.	−0·003	−0·002	−0·007*	−0·006	−0·001	−0·021	−0·007*	−0·021*
Superphosphate 4·5 cwt.	−0·008	0·002	−0·003	0·001	0·004	−0·005	−0·003	0·013
Sulphate of ammonia 2·0 cwt.	—	—	−0·003	0·004	—	—	−0·0c2	0·004
Standard error ±	0·003	0·002	0·002	0·004	0·009	0·011	0·004	0·008
Interaction K/salt	0·004	0·003	0·008*	0·001	0·008	0·008	0·006	−0·002
Standard error ±	0·004	0·003	0·003	0·005	0·015	0·019	0·006	0·011

Table VIII

% P in the dry matter

	Roots				Tops			
Experiment	1	2	3	4	1	2	3	4
Mean P %	0·291	0·157	0·210	0·161	0·163	0·135	0·189	0·194
Response to								
Salt 4·5 cwt.	0·022	0·026*	0·015	0·025**	−0.004	0·025*	0·008	0·024*
Muriate of potash 1·5 cwt.	−0·029*	−0·022	0·014	0·006	0·039*	−0·006	0·020*	0·013
Superphosphate 4·5 cwt.	0·018	0·055**	0·017	0·030**	0·037*	0·033*	0·016	0·051**
Sulphate of ammonia 2·0 cwt.	—	—	0·019	0·000	—	—	0·004	0·025*
Standard error ±	0·008	0·008	0·010	0·003	0·011	0·008	0·008	0·010
Interactions P/salt	−0·003	−0·013	−0·008	−0·016*	0·018	−0·004	0·011	−0·024
Standard error ±	0·013	0·014	0·014	0·005	0·020	0·014	0·013	0·014

reached 0·025% of P in both roots and tops. Superphosphate also increased phosphorus uptake in these experiments by amounts ranging up to 0·055%. There were frequent negative interactions between salt and superphosphate.

Discussion

Comparative effects of sodium and potassium chlorides

None of the soils could be described as deficient in available potassium and yield responses

to potassium were consequently small, a significant increase being obtained in only one experiment. Returns from the use of salt have been considerably greater in all the experiments except this one and in three cases there were large negative interactions between salt and potassium. Similar results have been found in a series of comparable experiments on kale and grass (to be published). It must therefore be concluded that salt has some value as a fertiliser for turnips even on soils which are not deficient in potassium. It cannot fairly be argued that larger responses would be obtained under conditions of potassium deficiency as many workers have claimed that a minimum level of potassium is necessary for the plant to be able to utilise sodium with advantage.

Salt and muriate of potash have directly opposing effects on the sodium uptake of turnips. Whereas salt stimulated the sodium content by about 50%, muriate of potash markedly reduced it by almost as much. Similar results (to be published) have been obtained for kale and grass. In every case the sodium content of the plant varied between very much wider limits than did the potassium, calcium, magnesium and phosphorus levels.

The practical significance of the depressive effect of potassium on sodium uptake is doubtful. It may however be relevant to draw attention to the fact that for sugar beet, which has a high sodium requirement, Pizer[30] has reported reductions in yield from the use of muriate of potash on soils which are already high in available potassium. For the same crop, Crowther[31] has also found small reductions of about 0·2 cwt. of sugar per acre (28 experiments) on soils with over 16 mg.-% of citric-soluble K_2O, from the application of muriate of potash. This should be contrasted with a mean increment of 1·2 cwt. of sugar (23 experiments) on soils containing 12–16 mg.-% of K_2O. It may therefore be possible that these small reductions are due to limitation of sodium uptake in the presence of applied potassium.

Salt did not have any appreciable influence on the potassium content of the crop. Both salt and muriate of potash depressed the calcium and magnesium contents, but by such small amounts as to be insignificant from the point of view of their mineral value to livestock.

Salt and superphosphate

Three of the soils were deficient in available phosphorus and there were consistent negative interactions between salt and superphosphate for the yields of roots. In Expts. 2 and 4 where soil phosphate was particularly low, there were striking differences in the responses of root yields to superphosphate in the presence and absence of salt, and vice versa (Table IX). Salt did not influence the yield of tops and the interactions here were irregular.

Table IX

Salt and superphosphate interactions

Expt.	Response to salt		Response to superphosphate	
	No superphosphate	With superphosphate	No salt	With salt
	Mean responses of roots, tons			
1	2·63	1·25	−0·05	−1·44
2	3·38*	0·08	8·14**	4·74**
3	1·30	0·16	1·94	0·80
4	4·48**	−0·74	7·63**	2·41**
	% P in roots			
1	0·025*	0·019	0·021	0·015
2	0·039*	0·013	0·068**	0·042*
3	0·023	0·007	0·025	0·009
4	0·041**	0·009	0·046**	0·014
	% P in tops			
1	−0·022	0·014	0·019	0·055*
2	0·029*	0·021*	0·037*	0·029*
3	−0·003	0·019	0·005	0·027
4	0·048**	0·000	0·076**	0·027*

Salt has increased phosphorus uptake in both roots and tops in all the experiments. Table IX also shows the effect of the large negative interactions between salt and superphosphate on

the phosphorus contents. Whilst Expts. 1 and 3 do not illustrate these effects so well, this work does nevertheless indicate that in certain circumstances salt enhances yields and phosphorus utilisation by turnips on soils of low phosphate status. Salt has been shown to be effective only in the absence of superphosphate.

Analyses of the soils from Expts. 2 and 4 after harvest did not reveal any higher readily soluble phosphorus status on the salt-treated plots. The larger growth on these plots could, of course, have depleted it.

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EFFECTS OF SALT AND OTHER FERTILISERS ON YIELD AND MINERAL COMPOSITION OF FORAGE CROPS. II.—Kale*

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Salt increased the yield of kale by about 1 ton/acre in five out of six experiments on soils which were not seriously deficient in available potassium. There were frequent negative interactions between salt and muriate of potash. Fertilisers, other than muriate of potash, did not materially influence the uptake of potassium. Salt and potash tended to depress the calcium and magnesium levels, but only slightly. Salt did not increase phosphorus utilisation although the soils were phosphate-deficient. Salt and ammonium sulphate greatly increased sodium uptake and muriate of potash markedly reduced it.

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Introduction

The suggestions that sodium may be a useful nutrient for Brassica crops, including kale, have been reviewed in Part I[1] together with the literature on the possible effects of salt on phosphorus uptake. There have, however, been no recorded experiments to investigate the effects of sodium on the yield and mineral composition of kale. Lehr[2] has quoted the results of a single trial in Ireland where salt reduced the yield of kale on a potassium-deficient soil, but stimulated it when potassium was also applied. Kale would appear to be a very suitable crop to investigate any effect which salt may have on the economy of soil potassium as amongst farm crops it removes as much total potassium from the soil as does the potato.

There has been a variety of experiments over a long period regarding the use of nitrogen on kale, the general conclusion being that 1 cwt. of ammonium sulphate produces about 1 ton of extra kale, the response being linear up to about 8–10 cwt. There are generally reductions in the content of dry matter and increases in that of crude protein. There seem to have been no investigations regarding the effects which nitrogen may have on the mineral composition of kale.

A search of the literature has revealed no records of experiments with phosphatic and potassic fertilisers on either the yield or mineral uptake of kale although compound fertilisers have been used at different rates.

Experimental

Six field experiments were carried out, two in each of the years 1954–1956. In the first four experiments the basic design was a $3 \times 3 \times 3$ layout to investigate all combinations of 0, 4 and 8 cwt. of ammonium sulphate, 0, 3 and 6 cwt. of superphosphate and 0, 1½ and 3 cwt. of muriate of potash. Salt at the rate of 4·5 cwt./acre was applied to a random half of each plot.

In 1956, the two experiments were in $2 \times 2 \times 2 \times 2$ designs. Each consisted of four blocks of eight plots with the NPK–salt interaction confounded between the two complete replicates of each site. The treatments were the presence and absence of 6 cwt. of ammonium sulphate, 4·5 cwt. of superphosphate, 4·5 cwt. of salt and 2 cwt. of muriate of potash.

The plot size (or half plot in Expts. 1–4) was 0·01 acre, each being 6 rows wide and 33 ft. in length. Edwards[3] and Halliday[4] have reported satisfactory results when using this size of plot, the latter obtaining coefficients of variation of between 7 and 8% for yields of dry matter. The fertilisers were applied by hand before the final seed bed cultivations. Normally about one week elapsed between fertiliser application and seed sowing. There were no adverse effects on germination. Half the ammonium sulphate dressings were reserved and applied as a top dressing after singling. Harvesting took place in November before the advent of frosty weather.

Sampling

The experimental areas were singled as well as possible, but there was some inevitable irregularity in the spacing. This naturally leads to the occurrence in any one plot of a small number of plants which are atypical from the remainder, being usually much smaller and stunted. Such plants do not contribute to the total weight in relation to their numbers and their composition might differ markedly from normal. They must therefore be rejected in sampling. Thus the plants sampled will not be truly random but be restricted to a selection from the more normal plants in each plot. Edwards[3] took six plants per plot and rejected any ' obviously atypical plants ' and Halliday[4] recommends that a fixed number (unspecified) of ' typical ' plants be taken.

There are obvious limitations to the number of plants which can be taken to form a composite sample in view of the bulk of material involved. The nature of the kale plant is such that fertilisers may alter the leaf/stem ratio and in view of their difference in composition it was decided that separate samples of leaves and stems for analysis must be obtained in the field.

In order to investigate sampling errors, ten groups of twelve plants from evenly manured areas of approximately 0·1 acre adjacent to Expts. 1 and 2 were selected and obviously atypical plants were ignored. The leaves were then stripped from the stems in the field. During this process, three leaves from each plant were collected separately. One was taken from the top

third, one from the middle portion, and one from the bottom third of the plant. These 36 leaves formed the laboratory sample for determination of dry matter and analysis. The bulked leaves and the stems from each of the groups of twelve plants were weighed separately to obtain the leaf/stem ratio. The twelve entire stems were chopped, reduced by quartering and dried for analysis.

Table I details the results of these investigations for each of the two areas and the analyses of the leaves and stems for the area adjacent to Expt. 1. The analyses were carried out according to the scheme described in Part I[1] and the data in Table I include the analytical errors.

Table I

*Sampling errors for kale**

(Ten samples of 12 plants)

	Experiment 1				Experiment 2			
	Min.	Max.	Mean	Standard deviation as % of mean	Min.	Max.	Mean	Standard deviation as % of mean
Fresh wt., lb.	30·0	34·5	31·9	6·22	27·0	33·5	30·3	7·11
Leaf stem ratio	1·19	1·63	1·37	9·40	1·30	1·70	1·50	9·99
% dry matter: leaf	11·2	11·9	11·5	2·38	12·1	12·7	12·4	1·70
stem	10·2	11·0	10·5	2·91	10·0	10·7	10·3	2·56
Analyses of dry matter (Expt. 1 only)								
% of dry matter	Leaf				Stem			
Na	0·235	0·300	0·275	7·74	0·285	0·350	0·322	7·02
K	2·10	2·80	2·47	8·23	3·05	3·85	3·50	7·14
Ca	2·04	2·70	2·22	9·63	0·80	0·96	0·89	7·61
Mg	0·110	0·146	0·130	10·15	0·132	0·162	0·152	8·82
P	0·265	0·315	0·292	5·38	0·275	0·325	0·299	7·22

* Including analytical error

Expressed on a % basis, all the errors fall between 7 and 10% of their respective means with the exception of the rather better results for determinations of dry matter. This order of magnitude is generally acceptable in this type of work. To reduce it further would have involved a disproportionate amount of work.

A further check on the validity of using twelve plants as representative of each plot was made. For each of the experiments, very good correlations were obtained between the weights of the 12-plant sample and the total harvested weight of each plot. The general practice was to take three plants from each of the four central rows of each plot to form the sample.

Results

In order to present the results of all six experiments in a comparable manner, the responses to 3 and 6 cwt. of superphosphate, 4 and 8 cwt. of ammonium sulphate and 1·5 and 3 cwt. of muriate of potash for the first four experiments have been averaged. The only discrepancy is that in Expts. 1–4 the potassium dressing is thus 2·25 cwt. compared with 2·0 cwt. in Expts. 5 and 6. Only those interactions for plant composition which were consistent or of significance have been tabulated. The remainder were both small and irregular.

Yields (Table II).—Salt gave increases in yield of approximately 1·0 ton of fresh kale in five of the six experiments. Three were significant ($P = 0·05$). Only one of the sites (Expt. 5) was on a soil considered very low in available potassium and only that one gave a large and significant response to muriate of potash. The only experiment (No. 6) which did not show a response to salt had the highest content of 1% citric-soluble potassium.

In only one case did the response to 2·25 cwt. of muriate of potash exceed that from 4·5 cwt. of salt, that being the soil lowest in available potassium (Table III). The responses were not well related to the 1% citric-soluble potassium, but none of the soils could be described as really deficient. Salt and muriate of potash normally interacted negatively.

Table II

Yields of fresh kale (tons)

Expt. No.		1	2	3	4	5	6
Mean yield		17·89	15·46	13·61	13·56	14·70	19·60
Response to							
Salt	4·5 cwt.	0·87*	0·95*	0·97*	0·93	1·06	−0·35
Muriate of potash	2·25 cwt.	0·20	0·83	0·50	0·44	3·40**	0·62
Superphosphate	4·5 cwt.	2·21*	0·99	0·19	4·22**	7·11**	1·28*
Sulphate of ammonia	6·0 cwt.	3·32**	4·85**	6·25**	5·52**	1·93**	2·02**
Standard error ±		0·50	0·80	0·68	0·50	0·63	0·40
Interactions							
NP		0·43	1·79	0·64	1·72*	1·69*	1·36*
NK		−0·24	−0·87	1·20	0·42	−1·04	−0·30
N/salt		1·18	−0·29	0·20	0·20	−0·22	−0·56
PK		−0·07	−2·51	−0·77	−2·06	1·82*	−0·77
P/salt		−1·37	0·29	0·20	0·05	−0·15	−0·55
K/salt		−1·43	−3·00**	−0·39	0·62	−0·32	0·28
Standard error ±		0·86	1·39	1·18	0·87	0·89	0·57

Table III

Soil analyses and responses to salt and muriate of potash

Expt. No.	1% citric-sol. K, mg.-%		Extra kale (tons) Potash	Salt
5	5·5	Low	3·40**	1·06
2	9·0	Low	0·83	0·95*
6	18·5	Satisfactory	0·62	−0·35
3	12·5	Satisfactory	0·50	0·97*
4	11·0	Low	0·44	0·93
1	8·5	Low	0·20	0·87*

The responses to superphosphate were well correlated with the soil analyses (Table IV). In contrast to the experiments with turnips there were no marked or regular negative P/salt interactions. There were, however, frequent and significant positive NP interactions.

Table IV

Soil analyses and responses to superphosphate

Expt. No.	1% citric-sol. P, mg.-%		Extra kale (tons)
5	3·5	Deficient	7·11**
4	4·5	Deficient	4·22**
1	5·0	Deficient	2·21*
6	7·5	Low	1·28*
2	10·0	Low	0·99
3	12·5	Low	0·19

Ammonium sulphate had large and significant (P = 0·01) effects on yield in all six experiments. The increases ranged from 2 to 6 tons.

In general, all treatments which stimulated yield tended to reduce the dry matter %, but not to such a degree as to obscure the significant responses.

Plant composition

Sodium (Table V).—Salt has had very large and consistent effects on sodium uptake, the increases being about equal in both leaf and stem and ranging from about 0·05 to over 0·40% of Na. In each case the increments were significant (P = 0·01) and were at least 50% of the mean sodium contents.

Ammonium sulphate had equally large effects. The responses were broadly correlated with the yield increments resulting from the use of nitrogen (Table VI).

Table V

% Na in dry matter

Expt. No.		1	2	3	4	5	6
Mean % Na	Leaf	0·383	0·329	0·270	0·477	0·348	0·130
	Stem	0·332	0·324	0·223	0·582	0·349	0·106
Response to							
Salt, 4·5 cwt.	Leaf	0·151**	0·164**	0·064**	0·343**	0·269**	0·048**
	Stem	0·121**	0·164**	0·073**	0·424**	0·199**	0·040**
Muriate of potash, 2·25 cwt.	Leaf	−0·233**	−0·213**	−0·246**	−0·266**	−0·292**	−0·043**
	Stem	−0·198**	−0·204**	−0·229	−0·316**	−0·266**	−0·037**
Superphosphate, 4·5 cwt.	Leaf	0·072*	0·030	0·029	0·030	0·124**	−0·001
	Stem	0·058*	0·014	0·002	−0·106*	0·115**	−0·008
Sulphate of ammonia, 6·0 cwt.	Leaf	0·147**	0·235**	0·218**	0·270**	0·065*	0·082**
	Stem	0·175**	0·226**	0·160**	0·282**	0·060	0·056**
Standard error ±	Leaf	0·016	0·015	0·032	0·014	0·028	0·009
	Stem	0·013	0·021	0·025	0·034	0·027	0·013
Interactions							
NK	Leaf	−0·074	−0·267**	−0·256**	−0·165**	−0·028	−0·005
	Stem	−0·106*	−0·223**	−0·232**	−0·212**	−0·020	0·003
K/salt	Leaf	−0·218**	−0·081	−0·069	−0·355**	−0·139**	0·007
	Stem	−0·169**	−0·102	−0·095	−0·222**	−0·266**	0·019
Standard error ±	Leaf	0·028	0·025	0·055	0·024	0·040	0·012
	Stem	0·023	0·036	0·044	0·060	0·038	0·018

Table VI

Effects of ammonium sulphate on yield and sodium uptake of kale

Expt. No.	Yield increase tons	Increase in % Na	
		Leaf	Stem
5	1·93**	0·065*	0·060
6	2·02**	0·082**	0·056**
1	3·32**	0·147**	0·175**
2	4·85**	0·235**	0·226**
4	5·52**	0·270**	0·282**
3	6·25**	0·218**	0·160**

Muriate of potash invariably reduced sodium levels and by amounts which were generally greater than the increments from salt and nitrogen.

Superphosphate had no consistent influence. In two experiments there were significant increases but these did not appear to be associated with the yield increments as other responsive sites showed no increase in sodium levels.

There were large and significant negative interactions between salt and potassium and between nitrogen and potassium on sodium uptake. The large effects of ammonium sulphate and salt were greatly reduced when muriate of potash was also applied.

Potassium (Table VII).—Salt has not altered the potassium content of either kale leaf or stem, the differences being generally in the order of only ±0·1% of K. Ammonium sulphate only increased potassium uptake markedly in Expt. 5 which was the only one to give a large yield increment to potash. Superphosphate tended to depress potassium levels in those experiments where it increased yields. Muriate of potash itself invariably raised the % K by amounts ranging up to 1%. There were no consistent or significant interactions.

Calcium (Table VIII).—The calcium content of kale has been singularly unaffected by fertilisers. Salt and muriate of potash tended to depress it, but by amounts which were generally under 0·1% (and frequently much less) in both leaf and stem. Neither ammonium sulphate nor superphosphate had any regular influence.

Magnesium (Table IX).—Both salt and potash reduced the amounts of magnesium in both leaf and stem. Only in one case, however, was the fall in magnesium significant and generally the reductions were under 0·01%. On the other hand, the tendency was for the ammonium sulphate to stimulate magnesium uptake, but again by only 0·01–0·02% Mg. There were no important interactions.

Table VII

% K in dry matter

Experiment		1	2	3	4	5	6
Mean % K	Leaf	1·64	2·44	2·63	2·62	2·19	3·17
	Stem	3·18	3·49	2·84	4·14	3·91	2·88
Response to							
Salt, 4·5 cwt.	Leaf	0·03	−0·12	0·10	−0·21	0·06	0·01
	Stem	−0·07	−0·03	0·10	−0·28	0·12	0·22*
Muriate of potash, 2·25 cwt.	Leaf	0·18	0·27**	0·74**	0·67**	0·88**	0·25*
	Stem	0·18	0·28	0·72**	0·66**	1·12**	0·16
Superphosphate, 4·5 cwt.	Leaf	0·21	−0·06	0·09	−0·32*	−0·41**	−0·14
	Stem	0·15	−0·15	−0·02	−0·34	−0·25**	−0·27**
Sulphate of ammonia, 6·0 cwt.	Leaf	−0·06	−0·20*	−0·13	0·20	0·24*	−0·05
	Stem	0·29	0·06	0·10	0·22	0·57**	0·01
Standard error ±	Leaf	0·11	0·05	0·14	0·08	0·11	0·11
	Stem	0·13	0·09	0·12	0·15	0·08	0·08

Table VIII

% Ca in dry matter

Experiment		1	2	3	4	5	6
Mean % Na	Leaf	2·20	2·19	2·33	2·65	2·95	2·76
	Stem	0·90	0·78	0·79	0·91	1·07	0·84
Response to							
Salt, 4·5 cwt.	Leaf	−0·05	−0·03	−0·02	−0·07	−0·27*	0·06
	Stem	0·02	0·00	−0·05*	0·01	−0·09	0·06
Muriate of potash, 2·25 cwt.	Leaf	−0·02	−0·03	−0·03	−0·11	−0·16*	−0·07
	Stem	−0·05	−0·05	0·01	−0·08	0·02	0·04
Superphosphate, 4·5 cwt.	Leaf	−0·08	−0·02	0·00	0·09	0·26*	0·07
	Stem	−0·05	−0·05	0·01	−0·08	0·02	0·04
Sulphate of ammonia, 6·0 cwt.	Stem	0·02	−0·02	0·07	0·03	0·07	0·08
	Leaf	0·02	−0·11	−0·01	0·02	0·10	−0·13
Standard error ±	Leaf	0·03	0·07	0·05	0·07	0·07	0·07
	Stem	0·03	0·01	0·03	0·04	0·04	0·02

Table IX

% Mg in dry matter

Experiment		1	2	3	4	5	6
Mean % Mg	Leaf	0·123	0·089	0·152	0·163	0·140	0·113
	Stem	0·152	0·133	0·161	0·165	0·200	0·119
Response to							
Salt, 4·5 cwt.	Leaf	−0·005	0·001	−0·011	0·002	−0·008	−0·001
	Stem	−0·012	0·000	−0·009	−0·004	−0·012	0·003
Muriate of potash, 2·25 cwt.	Leaf	−0·004	−0·012	−0·026*	−0·013	−0·004	−0·004
	Stem	−0·002	0·000	0·001	0·005	0·004	−0·004
Superphosphate, 4·5 cwt.	Leaf	−0·006	0·004	−0·015	0·022	0·012	−0·007
	Stem	−0·004	0·001	0·001	−0·007	0·004	0·001
Sulphate of ammonia, 6·0 cwt.	Leaf	−0·001	c·004	0·020*	0·015	0·002	0·009
	Stem	0·006	0·010*	0·003	0·012	0·015*	0·004
Standard error ±	Leaf	0·003	0·004	0·006	0·010	0·005	0·005
	Stem	0·005	0·003	0·007	0·009	0·005	0·005

Phosphorus (Table X).—Salt, potash and ammonium sulphate also had no regular or significant effect on the phosphorus content of either leaf or stem. Superphosphate stimulated phosphorus absorption in all six experiments. These increases do not appear to be related to the yield response to superphosphate and reached significance only in the stems of two experiments. There were no consistent interactions.

Table X

% P in dry matter

Experiment		1	2	3	4	5	6
Mean % P	Leaf	0·289	0·310	0·180	0·212	0·234	0·288
	Stem	0·320	0·360	0·210	0·264	0·280	0·388
Response to							
Salt, 4·5 cwt.	Leaf	0·010	0·014	−0·008	−0·003	−0·003	0·033
	Stem	0·019	0·010	−0·021	−0·003	0·022	0·017
Muriate of potash, 2·25 cwt.	Leaf	−0·011	0·012	0·008	0·10	−0·008	0·018
	Stem	−0·018	0·006	0·015	−0·011	−0·006	−0·016
Superphosphate, 4·5 cwt.	Leaf	0·015	0·027	0·001	0·013	0·004	0·014
	Stem	0·029*	0·028	0·007	0·056*	0·018	0·011
Sulphate of ammonia, 6·0 cwt.	Leaf	0·010	0·022	0·001	0·007	−0·008	0·030
	Stem	0·014	0·003	−0·004	0·009	−0·011	0·011
Standard error ±	Leaf	0·007	0·010	0·003	0·007	0·009	0·017
	Stem	0·009	0·010	0·009	0·014	0·010	0·007

Discussion

Comparative effects of sodium and potassium chlorides

Salt increased the yield of fresh kale in five of the six experiments and only one of the sites could be described as so low in available potassium that a large and significant response was obtained to muriate of potash. The five increments from 4·5 cwt. of salt ranged from 0·87 to 1·06 tons whereas 2·25 cwt. of muriate of potash raised the yield by about 0·5 tons with one exceptional rise of 3·4 tons. It can thus be concluded that salt may increase the yield of kale at least under the conditions of these experiments, i.e., on soils which are not unduly deficient in potassium. The frequent negative K–salt interactions may be indicative of possibly greater responses from salt on soils of lower potassium status.

Both salt and potash tend to depress the uptakes of calcium and magnesium, but by only about 0·1% Ca and 0·01% Mg. They do not therefore materially influence the value of kale as a source of these minerals for livestock.

Salt and superphosphate

In contrast to the experiments with turnips (Part I[1]) there were no marked negative interactions between salt and superphosphate for either the yield or phosphorus uptake of kale in spite of the fact that significant yield responses to phosphate were obtained in four of the six kale experiments. Salt had no regular effect on phosphorus uptake.

Sodium uptake

Salt invariably increased the amount of sodium in the whole plant by about 50%. In contrast, there were rather greater depressions when muriate of potash was applied. These results are entirely analogous to those obtained with turnips (Part I[1]).

Very striking increases in the absorption of sodium followed the use of ammonium sulphate and it has been shown (Table VI) that these could be correlated with the responses in yield. Similar results (to be published) have been found with grass. With turnips on the other hand, nitrogen affected neither yields nor sodium uptake. Enhanced sodium uptake thus appears to be governed, in part, by other factors which themselves increase yield. In the case of kale (and grass) nitrogen is the dominating influence, but in turnips it may be phosphate. This may be due to a definite need of the plant or, in a sense, be accidental. Increased growth is normally associated with greater water uptake and there may be secondary influences in root size and permeability. Scharrer & Jung[5] have recently published the results of some pot experiments with maize to investigate the effects on mineral composition of sodium, potassium and calcium supplied as different anions. They found that only anions important in plant nutrition promote the penetration into plants of cations such as sodium for which there is normally a small requirement. The uptake of potassium, due to its essential nature, was unaffected by the anion. The large amount of work concerning the uptake of sodium from experiments using sodium nitrate must therefore be interpreted with caution as a good deal of the increase in plant sodium may be due to the nitrate ion.

These experiments and those with turnips (Part I[1]) and grass (to be published) indicate that the sodium content of crops varies within very wide limits relative to those of potassium, phosphorus, calcium and magnesium. Only the first two are likely to be altered significantly by the use of appropriate fertilisers.

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AMINO-ACID COMPOSITION OF HERRING (*CLUPEA HARENGUS*) AND HERRING MEAL. DESTRUCTION OF AMINO-ACIDS DURING PROCESSING

By GJERMUND BOGE*

The complete amino-acid composition has been determined for herring, herring meal and intermediates in the production of the latter. The values found for herring meal agree fairly well with earlier published data. No destruction seems to take place under processing at 'optimal' conditions, while condensing of press water at extremely high temperature causes severe damage to several amino-acids. Destruction of amino-acids is also pronounced in heat-damaged herring meal.

Introduction

Only a few papers report studies of the complete amino-acid composition of herring meal, although Connell & Howgate[1] report values for herring fillets. No complete analyses, however, are recorded with regard to the contents in fresh herring and some typical intermediates obtained during production of herring meal (i.e., press cake and press water). The present paper reports studies by microbiological methods of the amino-acid composition of herring, intermediates and by-products obtained during the manufacture of herring meal. Proper samples have been selected to make possible a study of the effect of heating on the amino-acid contents during processing.

Experimental

Samples

Herring (Clupea harengus).—An average sample from about 500 hl. of fresh herring caught off the West coast of Norway in January was used in the production of the samples following.

Herring press-cake.—An average sample of press cake from about 50 hl. of herring. This was dried to afford *herring press-cake meal.*

Herring whole meal.—This is a mixture of the press cake and its press water added in theoretical amounts after separation of the fat. For use, the press water was initially condensed at about 120°, finally at about 105° to a dry matter content of approximately 40%.

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This process was carried out in a pilot plant under ' optimal ' conditions. The press cake was flame-dried at an inlet temperature of about 400° and an outlet temperature of about 80°.

Herring press-water.—An average sample of herring press-water was obtained from the production in an ordinary factory and the fat separated.

Herring solubles.—This press water was condensed in a triple condenser at 156°, 143° and 133° in the three stages, respectively, to a dry matter content of approximately 30%.

Spontaneously heated meal was taken from a store. The meal was dark brown in colour and was considered of a bad quality.

Preliminary treatment of samples

The samples of press water and solubles were heated to 50° and the fat removed after centrifugation. The press water was then condensed in vacuum at 45° to a dry matter content of approximately 30%.

The other samples were also dried in vacuum at 45° and extracted with light petroleum (b.p. 50–70°) in a Soxhlet extractor. Finally the extracted samples were dried in vacuum at room temperature over calcium chloride overnight.

Methods of analysis

Dry matter was determined by drying in an oven for 4 h. at 105°, except for the samples of press water and solubles which were dried for 18 h.

Ash was determined by ignition of the dried samples in an oven for 18 h. at 540°.

Protein was calculated from the Kjeldahl nitrogen value multiplied by the factor 6·25.

Amino-acids.—These were determined by microbiological methods (see Table I) and potentiometric titration of the lactic acid produced after incubation for 72 h. An automatic dispenser titrator (Cannon type) was employed, and all assays were carried out at a total volume of 2·0 ml. per tube. In the main, the procedures in the literature cited were followed, but certain modifications were made, mainly as regards the temperature, and in one case the pH of the medium has been adjusted to 6·0 (proline assay). These modifications were found to give better standard curves. In all assays a sterilisation time of 5 min. at 120° was employed. The test organisms used were obtained from the American Type Culture Collection and maintained as described by Barton-Wright.[2]

Table I

Experimental conditions for the microbiological assays

Amino-acid	Medium	Test organisms	Standard curve, μg./2 ml.	Incubation temperature, °C
Leucine	Barton-Wright[2]	*Lactobacillus plantarum* (8014)	0–20	30
Isoleucine	,,	,, ,, ,,	0–20	30
Valine	,,	,, ,, ,,	0–20	30
Lysine	,,	*Leuconostoc mesenteroides* (8042)	0–40	37
Phenylalanine	,,	,, ,, ,,	0–10	37
Histidine	,,	,, ,, ,,	0–5	37
Aspartic acid	,,	,, ,, ,,	0–40	37
Proline	,, *	,, ,, ,,	0–20	37
Serine	,,	,, ,, ,,	0–15	37
Glycine	,,	,, ,, ,,	0–20	37
Glutamic acid	,,	*Lactobacillus plantarum* (8014)	0–35	37
Methionine	,, **	*Leuconostoc mesenteroides* (8042)	0–10	37
Tyrosine	,, **	,, ,, ,,	0–12	30
Alanine	,,	*Leuconostoc citrovorum* (8081 & 8082)	0–50	37
Arginine	,,	*Streptococcus faecalis* (8043)	0–15	37
Threonine	,,	,, ,, ,,	0–15	37
Tryptophan	Kuiken *et al.*[3]	,, ,, ,,	0–3	37
Cystine	Shockman *et al.*[4]	*Leuconostoc mesenteroides* (8042)	0–10	37

* pH of medium adjusted to 6·0

** Medium supplemented with 0·01 μg. of biotin per ml. (double strength)

Each amino-acid was determined in two separate assays, carried out on two separate hydrolysates of the samples at different times. In each series, six assay levels were used and recovery experiments according to Snell's method[5] were carried out for all amino-acids. Recoveries ranged from 95 to 106%, with a mean value of about 102% for all acids except cystine (recovery 106–113%, mean value 110%). Purity of the acids used was checked by paper chromatography (Dent[6]), and acids used as standards were dried at room temperature in a vacuum desiccator over calcium chloride.

Hydrolysates for tryptophan assay were prepared by the method of Kuiken *et al.*[3] After preliminary experiments, the method of Alexander *et al.*[7] was chosen for the preparation of hydrolysates for assay of cystine. Acid hydrolysates used for the determination of the remaining amino-acids were prepared by the method given by Barton-Wright,[2] but hydrolysis was for 10 h.

Results

The content of crude protein (N × 6·25), ash and dry matter of the samples are given in Table II. Amino-acid compositions calculated as g. of amino-acid per 100 g. of protein are recorded in Table III.

Table II

Analyses of the defatted samples

Sample	Dry matter, %	Protein (N × 6·25), %	Ash %
Herring	90·5	76·2	10·5
Herring press-cake	95·0	80·8	10·6
Herring press-cake meal	95·1	79·6	10·5
Herring whole meal	95·3	80·8	11·2
Herring press-water	32·05	27·3	5·2
Herring solubles	30·35	25·1	5·36
Spontaneously heated meal	95·8	85·2	10·2

Table III

Amino-acid composition of herring etc. samples

Amino-acid	g. amino-acid per 100 g. of crude protein (N × 6·25)						
	Whole herring	Press cake	Press-cake meal	Whole meal	Spontaneously heated meal	Press water	Solubles
Arginine	7·14	8·15	8·10	7·86	7·10	5·38	5·22
Histidine	1·87	2·03	2·10	1·84	1·46	1·21	0·69
Isoleucine	6·20	6·75	6·70	6·12	5·55	1·98	1·91
Leucine	7·14	7·45	7·60	6·85	6·30	3·29	3·24
Lysine	8·34	9·05	9·06	8·18	5·72	4·57	4·31
Methionine	2·56	2·65	2·72	2·49	2·22	1·32	1·36
Phenylalanine	3·57	3·85	3·87	3·55	3·26	1·61	1·54
Threonine	4·1	4·07	4·19	4·0	3·52	2·24	2·07
Tryptophan	0·78	0·82	0·81	0·72	0·52	0·156	0·099
Valine	5·38	5·74	5·85	5·33	4·74	2·57	2·46
Tyrosine	3·0	3·27	3·30	2·89	2·55	0·72	0·68
Cystine	1·4	1·6	1·6	1·3	0·95	0·42	0·08
Glycine	6·31	5·4	5·66	6·28	5·59	10·2	10·9
Alanine	7·64	7·71	7·45	7·45	7·04	7·29	7·89
Aspartic acid	9·42	9·90	9·82	9·10	8·80	4·96	4·92
Glutamic acid	11·44	12·05	12·03	11·75	10·6	7·73	7·53
Proline	4·23	4·34	4·32	4·6	4·1	4·6	4·8
Serine	4·1	4·5	4·63	4·15	4·05	2·9	—

Discussion

The values for the amino-acid content of herring press-cake meal agree well with results reported in the literature.[8] As the recoveries of amino-acids added to the hydrolysates were satisfactory, the values presented are considered to give a fairly true picture of the amino-acids composition of the materials investigated, although the values for threonine and serine may be somewhat low owing to losses during the hydrolysis.[1, 9]

A comparison of the data for press cake and press-cake meal and for the whole herring and the whole herring meal show that no destruction seems to have occurred under the processing conditions described.

The spontaneously heated meal was, as mentioned above, not from the same raw material as the remaining samples. Fortunately, however, a determination of lysine had been carried out in fresh meal from the same production, when a figure was obtained agreeing with the values found for the present samples of herring whole meal. It may therefore be assumed that the spontaneously heated meal is comparable to herring whole meal and that the spontaneous heating led to destruction of the amino-acids. Compared with herring whole meal, the destruction during spontaneous heating is most pronounced for lysine, tryptophan, cystine and histidine, with respectively 30, 27, 27 and 20% loss. For most of the remaining amino-acids investigated the values are approximately 10% lower than for the fresh meal. With regard to the protein quality of the spontaneously heated meal, this has been investigated by Laksesvela,[10] who found much reduced biological and feeding quality for this meal.

The temperature and the time of drying during the processing are factors which must be taken into consideration. A comparison of the values for herring press-water and for condensed herring solubles, clearly shows that an appreciable destruction of some of the amino-acids may take place during evaporation. The cystine is nearly completely destroyed, while the losses of histidine and tryptophan amount to 43 and 36% respectively. For lysine it is interesting to note that while the destruction in the spontaneously heated meal is 30%, condensing the press water causes a loss of only 5%. The remaining amino-acids seem to be quite stable during this process.

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VARIATION IN THE NITROGEN CONTENT OF TEA LEAVES

By D. N. BARUA and S. B. DEB

Data are presented showing the relationship of the nitrogen content of tea leaves to the level of nitrogenous manuring, to the position in plucked and unplucked shoots, and to various genetic and environmental factors. In carrying out comparative studies of the chemical constituents of tea leaves it is suggested that sampling should be confined to a particular leaf in a specified position on the stem. Samples drawn from plucked shoots should consist of the second leaves of comparable shoots taken from bushes growing in the inner rows of a plot.

Introduction

The past records of this Station contain many attempts to correlate the nitrogen content of plucked tea shoots with the yield and market value of the processed tea. Enhanced yield, due to nitrogenous manuring, is correlated with the nitrogen content of the shoot (Table I) and there have been indications that an increase in the nitrogen content of shoots tends to reduce the value of a tea.[1]

A detailed investigation of the relationships of leaf nitrogen to yield and quality of tea would require estimations of proteins, amino-acids and caffeine. Before undertaking such a task it seemed to be desirable first to study the variability of total nitrogen content in different clones at various stages of development and under diverse conditions of growth and to standardise a procedure for sampling leaves for analysis based upon the findings of this preliminary investigation.

Materials and methods

In northern India, tea bushes are plucked to a flat, horizontal surface, normally at weekly intervals. Shoots which project above the plucking level by two leaves or more, and dormant shoots (the terminal bud in the dormant state) with only one leaf above the level are harvested as the tea crop. While there are minor modifications to this system, the harvested crop always consists of a mixture of shoots at various stages of development. Fig. 1 is the diagram of a plucked tea shoot with three leaves and a growing terminal bud.

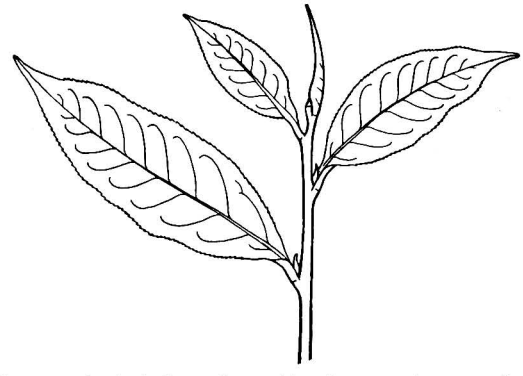

FIG. 1.—*A plucked tea shoot with a long growing apex (bud)*

The stem portions between first and second ,and second and third leaves, are designated stalk 1-2, and stalk 2-3, respectively, in Table II

Tea shoots pass through alternating states of growth and dormancy as described by Wight & Barua.[2] The amount of growth between two successive states of dormancy is termed a flush. The unplucked shoots used in the present experiments (cf. Table III) consisted of two flushes, having five leaves in each flush.

Leaves from four sources were used in these investigations :

(*a*) A population of tea of the Assam variety (*Camellia sinensis* var. *assamica*) raised from

a seed source (*bari*) known as ' Mesai Manipuri '. This block was used for a NPK manuring trial. Bushes without shade, spaced 4·5 feet triangular, 23 years old.

(*b*) A small 25-bush plot of clone 16/10/22 (var. *assamica*). Bushes without shade, spaced 4·5 feet square, 12 years old.

(*c*) A plot of 95 bushes of clone 19/10/3 (var. *assamica*). Bushes without shade, spaced 4·5 feet square, 8 years old.

(*d*) Five morphologically distinct bushes ranging from var. *assamica* through various intergrades to var. *sinensis*. Bushes without shade, spaced 5 feet square, 28 years old.

It is to be noted that in either system of planting, square or triangular, most bushes are surrounded by other bushes, excepting the peripheral rows, adjacent to a main drain or pathway.

Total nitrogen was estimated by the micro-Kjeldahl method following the procedure of Ma & Zuazaga.[3]

Results expressed on area basis were obtained by analysing circles of leaf tissue cut out with a Ganong leaf punch.

Results

Dose of fertiliser, leaf nitrogen and yield

The results of 15 successive years' manuring of the Mesai Manipuri block of unshaded tea with sulphate of ammonia have consistently shown a linear response of the increase in yield to nitrogenous manuring up to a level of 135 kg. of N per hectare. Table I shows the yields recorded in the year 1944. There is also a linear relationship (up to 179 kg. of N per hectare) between the % N in the plucked shoots and the amount of nitrogenous manure applied. The yield is significantly correlated ($r = 0{\cdot}98$; $P < 0{\cdot}001$) with the % of N in the shoots.

Table I

Dose of fertiliser, yield and % N in plucked shoots of the Mesai Manipuri plots, for the year 1944

N supplied as fertiliser, kg./ha.	Yield of made tea, kg./ha.	% N on dry weight of shoots
0	563	4·67
45	920	4·89
90	1369	5·02
135	1750	5·16
179	1798	5·32
224	1840	5·30
L.S.D. at P = 0·05	142	0·18

Nitrogen distribution in the component parts of plucked shoots

As shown in Table II the N content of both leaf and stalk decreases with age. These results also show a considerable difference between dates.

Table II

% *N on dry matter in component parts of shoots plucked from clone* 19/10/3

Components of shoot	Date			Average
	Mid June	Early July	End July	
Bud	6·99	7·71	7·17	7·29
1st leaf	5·54	5·13	5·71	5·46
2nd leaf	5·41	4·88	5·07	5·12
3rd leaf	5·23	4·78	5·03	5·01
Stalk, 1–2	4·01	4·75	4·48	4·41
Stalk, 2–3	3·96	4·73	4·43	4·37

Nitrogen in successive leaves of unplucked shoots

The % of N on a dry weight basis decreases progressively from the first leaf to the tenth (Table III) and % N per unit area of leaf decreases more slowly from the first leaf to the fourth after which it shows no further change. The variations in leaf area agree with the previous observations of Wight[4] that the fifth leaf of a growing tea shoot is fully expanded.

Table III

Area and composition of successive leaves on unplucked shoots of clone 16/10/22

Leaf position	Leaf area, sq. cm.	Dry weight (mg.) per sq. cm. leaf area	N on dry weight, %	N (mg.) per sq. cm. leaf area	N in entire leaf, mg.
1	6·6	5·28	6·27	0·33	2·18
2	16·9	5·62	5·30	0·30	5·07
3	37·5	5·99	4·68	0·28	10·50
4	51·3	6·42	4·20	0·27	13·85
5	51·0	6·50	4·18	0·27	13·77
6	53·0	6·57	3·91	0·26	13·78
7	51·5	6·61	4·02	0·26	13·39
8	46·1	6·62	3·96	0·26	11·99
9	41·3	6·54	3·82	0·25	10·32
10	34·9	6·43	3·69	0·24	8·38

Choice of sample

The data in Tables II and III show that the composition of a leaf varies with its position on the stem. If only one leaf is to be used for comparisons of shoots it might appear that the fourth or fifth leaf would be the best choice but these leaves are not normally used for manufacture. The choice therefore falls upon the second leaf, as this leaf always forms part of shoots plucked for manufacture and constitutes a considerable fraction of the total bulk of these shoots.[5]

In the following results describing the effects of genetic and environmental factors on nitrogen content, the samples used consisted entirely of the second leaves separated from the shoots.

Genetic and environmental factors

Kind of tea.—Tea bushes can be arranged subjectively in a series of intergrades ranging from the extreme form of the var. *assamica* to the extreme form of the var. *sinensis*. The values for % N (dry weight basis) obtained for the second leaf of varieties closer to *assamica* and *sinensis* respectively were 4·95 and 5·80, and for intergrades progressively approaching *sinensis* 5·20, 5·50 and 6·23. The number of replicates examined was insufficient for statistical significance but there would appear to be a tendency for N content to vary with the kind of tea.

Position of bush in plot.—Leaf from bushes in the outside rows of a plot (i.e., adjacent to a drain or pathway) has a significantly lower N content than leaf plucked from rows within the plot (Table IV). Each figure in this table is a mean of eight separate estimations, each on a different date. In this experiment there was no significant difference between bushes in the inner rows. This is not always the case as exemplified in Table V. The significant differences in this latter case may be the result of a soil gradient.

Date variance.—The results presented in Table VI show significant differences in N content associated with date of plucking.

Position and size of shoots on bush.—The position of the shoot on the bush has no significant effect on the N content (Table VII), while there is a tendency for the second leaf to have a higher N content when it forms part of a shoot consisting of two leaves and a bud (Table VIII).

Size of terminal bud.—Short or long terminal buds are defined as being less than half or more than half the length of the first leaf. The size of the bud does not appear to affect the N content of the second leaf (Table IX).

Size of second leaf.—The total N content of leaves of various ages has been shown to be proportional to area. The same relation holds good for the second leaves of unequal sizes of clone 19/10/3. % N on dry weight shows a tendency to decrease with the increasing size of the leaf ($r = -0{\cdot}602$; $P < 0{\cdot}01$). This seems to be an age effect (cf. Table III). N per unit area, however, does not differ significantly between second leaves of various sizes (see Table X).

Time of day.—Samples of leaves are usually collected during the middle hours of the day. To ascertain if leaf-N changes during these hours, second leaves of clone 16/10/22 were sampled at 11 a.m. and 2 p.m. Analyses (Table XI) showed that the difference in the N contents of these two samples is not significant.

Table IV

N in 2nd leaves of inner and outer rows of bushes of clone 19/10/3

Position of the bush in the plot	Bush No.	N on dry weight, %	N (mg.) per sq. cm. leaf area
Inner rows	B 8	5·12	0·292
	B 9	5·28	0·301
	C 4	5·30	0·297
	C 8	5·32	0·294
	D 9	5·46	0·308
	D 10	5·34	0·289
	mean	5·31	0·297
Outer rows	A 1	5·01	0·277
	A 8	4·73	0·264
	E 4	4·98	0·277
	E 8	4·99	0·274
	mean	4·93	0·273
L.S.D. at P = 0·05		0·37	0·022

Table V

% *N in 2nd leaves of bushes along the diagonal of a plot of clone* 16/10/22

Position of the bush		N on dry weight, %
Row no.	Bush no.	
A	5	5·38
B	4	5·54
C	3	5·74
D	2	5·78
E	1	5·92
L.S.D. (P = 0·05)		0·36

Table VI

% *N in the 2nd leaves of clone* 16/10/22 *on different dates*

Date	N on dry weight, %
22nd July	5·19
3rd August	4·85
17th August	6·28
11th October	6·36
L.S.D. at P = 0·05	1·50

Table VII

N in 2nd leaves of centre and side shoots of clone 16/10/22

Position of the shoot on the bush	N on dry weight, %	N (mg.) per sq. cm. leaf area
Centre	6·48	0·333
Side	6·22	0·336
L.S.D. at P = 0·05	0·35	0·018

Table VIII

N in 2nd leaves of shoots carrying two and three leaves (Clone 16/10/22)

Size of shoots	N on dry weight, %	n (mg.) per sq. cm. leaf area
With 3rd leaf	5·40	0·200
Without 3rd leaf	5·50	0·208
L.S.D. at P = 0·05	0·10	0·010

Table IX

% *N in 2nd leaves of shoots with short and long buds* (Clone 16/10/22)

Repeat	Short bud	Long bud
1	5·42	5·44
2	3·87	3·80
3	5·58	5·79
4	5·06	5·12
5	4·00	4·06
Mean	4·79	4·84

L.S.D. at P = 0·05 : 0·12 for mean

Table X

N in 2nd leaves of various sizes (Clone 19/10/3)

Area of 2nd leaf, sq. cm.	N on dry weight, %	N (mg.) per sq. cm. leaf area	N per leaf, mg.
6·0–6·9	5·28	0·300	1·891
7·0–7·9	5·76	0·329	2·320
8·0–8·9	5·63	0·286	2·424
9·0–9·9	5·75	0·287	2·748
10·0–10·9	5·54	0·303	3·078
11·0–11·9	—	—	—
12·0–12·9	5·71	0·310	3·603
13·0–13·9	5·55	0·314	4·076
14·0–14·9	5·36	0·296	4·226
15·5	4·63	0·289	4·480

Table XI

% *N in 2nd leaves of clone* 16/10/22 *at different hours of the day*

Date	N on dry weight at %	
	11 a.m.	2 p.m.
18th July	6·60	6·48
23rd ,,	4·34	4·28
24th ,,	4·24	4·20
28th ,,	4·97	5·00
29th ,,	5·11	5·12
Mean	5·05	5·02

Discussion

So long as the tea leaf is expanding the N content per unit area decreases progressively; once the leaf is fully expanded there is little or no further change in N content per unit area. On a weight basis, however, there is a progressive fall in the N content of ageing leaves, which is attributed to a gradual increase in non-nitrogenous matter.

The size of shoots, used for the manufacture of tea, varies from bush to bush and the proportions of the various component parts of the shoot are altered by changes in the environment.[5] This adds to the heterogeneity of the material which is already apparent from analyses of the individual portions of the shoot. The chemical analysis of plucked shoots, therefore, demands very careful sampling, especially in the case of populations raised from seed where leaf-N has been found to show great variations between bushes. However, if a direct comparison between fresh shoots and processed tea is required, the sample must consist of whole shoots. The number of replicates must therefore be large enough for the standard error to be reasonably low.

For comparisons of clones and other populations and reactions to changes in environment it is claimed that samples consisting of one particular leaf are to be preferred to whole shoots. The sample is then of greater homogeneity, and a sample of the same weight can include representatives of a greater number of bushes in the population. The precision can be further increased by confining the sample to leaves of uniform size, drawn from equally sized shoots from bushes in the inside rows of a plot. The sampling must be repeated on at least five different dates.

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THE STABILISING EFFECT OF PIPERONYL BUTOXIDE ON PYRETHRINS EXPOSED TO ULTRA-VIOLET LIGHT

By J. M. DONALDSON and J. H. STEVENSON

A mixture of the synergist piperonyl butoxide with pyrethrins is not significantly more toxic to adult moths than are pyrethrins alone. The reported stabilisation in ultra-violet light of pyrethrins by piperonyl butoxide could therefore be investigated biologically without the complication of synergistic activity. Stabilisation was not demonstrated.

Introduction

In spite of their relatively high cost as compared with other insecticides, the insecticidal constituents of pyrethrum are still used extensively in certain fields of insect pest control. Their low mammalian toxicity and rapid knockdown action make them particularly suitable for use in connexion with stored food products and public health spray programmes. The high cost of pyrethrins has led to the extensive use of synergists which enhance the toxicity of the insecticide and so enable smaller concentrations to be used in formulations.

For the sake of brevity the insecticidal constituents of the extract of pyrethrum flowers are here referred to as pyrethrins.

The instability of pyrethrins, especially on exposure to light, makes them less suitable than many synthetic insecticides for establishing residual insecticidal films. Blackith[1] demonstrated that this instability in the presence of light was due mainly to the ultra-violet, rather than the visible, wavelengths. Nasir[2] showed that the maximum absorption of light by pyrethrins was reached at about 2300 Å.

It has been claimed that besides enhancing the toxicity of pyrethrins, synergists may also reduce the rate at which they are broken down. The evidence on this point is contradictory. Blackith[3] concluded, after reviewing work by Hewlett and by Nash as well as his own, that the stabilisation of pyrethrins by the synergist piperonyl butoxide, during exposure to light under three conditions, including irradiation by ultra-violet light, was negligible if indeed it occurred at all. Dove[4] claimed to have demonstrated stabilisation in tests in which panels treated with pyrethrins and piperonyl butoxide remained toxic to flies for several months. This observation is consistent with the view that the concentration of the insecticide on the treated panels was such that the breakdown of pyrethrins that occurred during the period of the experiment was not sufficient to reduce the toxicity of the deposit to the flies. Phipers & Wood[5] have reported that piperonyl butoxide stabilises the pyrethrins when a solution is irradiated with ultra-violet light. They used the sulphur-colour colorimetric method for analysing pyrethrins, which was adapted for estimating true pyrethrins in degraded material.

In view of the difficulty of establishing, by chemical analysis, small changes in concentration of the insecticidal constituents of extracts of pyrethrum, it was decided to investigate this subject by means of a biological method of assay. The solutions of piperonyl butoxide and degraded pyrethrins were tested by topical application on adults of *Ephestia cautella* (Walk.) using the micro-burette method of Kerr.[6]

Brooke[7] has described the very low level of synergistic activity shown by piperonyl butoxide/pyrethrins mixture towards *Ephestia elutella* (Hb.). Stevenson,[8] using *Plodia interpunctella* (Hb.) and *Ephestia cautella*, failed to demonstrate any significant enhancement of the toxicity of pyrethrins films by the addition of piperonyl butoxide.

Because the synergistic effect of piperonyl butoxide on the toxicity of pyrethrins to *E. cautella* adults is negligible, this insect is particularly suitable for biological assays designed to demonstrate any stabilising effect the synergist may have during irradiation of pyrethrins with ultra-violet light. Results obtained from these assays would not be confounded by the synergistic action of piperonyl butoxide.

Experimental

The pyrethrins were obtained as a concentrate containing 20% of active material. To a measured volume of concentrate was added an equal volume of absolute alcohol to precipitate the oleoresins. This 10% solution was then stored in a dark refrigerator to prevent deterioration due to light and heat. Small volumes of this solution were diluted for use by the addition of odourless kerosene.

Irradiation of the solutions was carried out by means of a G.E.C. 125-W. ' Osira ' mercury vapour discharge lamp from which the outer glass cover had been removed in the manner described by Blackith,[1] so that it emitted ultra-violet rays. The output of light energy from the lamp when used in this way was about 15% of the wattage at 2650 Å, 10% at 3126 Å, 4% at 3022 Å : the total output below 3000 Å was about 5%, and between 4000 and 6000 Å was 50%. (This information was supplied by the General Electric Co.)

This lamp was contained in a black wooden box and the emergent rays were roughly collimated by interposing two asbestos sheets, each pierced with a circular hole, between the lamp and the material being irradiated. These sheets served to protect the solution from indirect heating. Two ml. of the solutions of pyrethrins in odourless kerosene were placed in an open 7-cm. Petri dish situated below the lamp and irradiated for 20 min.

The moths were lightly anaesthetised with carbon dioxide and each of them was dosed by the method of Kerr with 0·05 μl. of solution, which was always applied as nearly as possible to the dorsal thorax of the moths. They were then enclosed on clean filter-papers for 2½ h.

at a constant temperature and humidity (approximately 25° and 70% R.H.). At the end of this time, the results were assessed by ' touching ' each moth lightly with a blunt wire and if it did not respond by moving about in a normal, co-ordinated manner, it was considered to be knocked down.

Series of doses of pyrethrins and of pyrethrins plus piperonyl butoxide were applied topically to *E. cautella* adults (Table I). Statistical analysis of these results confirmed Stevenson's findings that the positions of the two regression lines for treatment in the presence and absence of the synergist cannot be distinguished, even though in this case a much higher ratio of synergist to pyrethrins than is usual (5 : 1 as in Tables III and IV) was used in the tests shown in Table I. This coincidence of the lines verified that irradiated pyrethrins could be analysed by a biological method that was not confounded by synergistic activity.

Table I

Topical application to Ephestia cautella *adults of doses of* 0·05 *µl. of solutions of pyrethrins and of pyrethrins plus piperonyl butoxide*

Concn. of pyrethrins, %	Concn. of piperonyl butoxide, %	Mortality Total	Mortality %*	Concn. of pyrethrins, %	Concn. of piperonyl butoxide, %	Mortality Total	Mortality %*
0·09	0	26/30	86	0·09	4·5	28/31	89
0·06	0	24/30	78	0·06	3·0	25/29	84
0·04	0	22/29	74	0·04	1·5	21/30	67
0·02	0	19/30	59	0·02	1·0	20/31	61

* Correction made for mortality of 3/31 in control with neither pyrethrins nor piperonyl butoxide

χ^2_2 for regression (pyrethrins only) = 1·08 ; $0{\cdot}7 > P > 0{\cdot}5$
χ^2_2 for regression (pyrethrins plus synergist) = 1·10 ; $0{\cdot}7 > P > 0{\cdot}5$
χ^2_6 for regression (combined results) = 1·12 ; $0{\cdot}99 > P > 0{\cdot}98$

When *E. cautella* adults were treated with a series of doses of irradiated and non-irradiated pyrethrins (Table II) a reduction of about 20% in the knockdown of the moths dosed with the irradiated solution was observed. A similar result was obtained (Table III) when adult moths were treated with a series of irradiated and non-irradiated solutions of pyrethrins plus piperonyl butoxide.

Table II

Topical application to Ephestia cautella *adults of doses of* 0·05 *µl. of solutions of irradiated and non-irradiated pyrethrins*

Concn. of pyrethrins, %	Mortality Total	Mortality %*	Concn. of pyrethrins, %	Mortality Total	Mortality %*
Non-irradiated			Irradiated		
0·09	25/30	82	0·09	14/31	43
0·06	18/30	59	0·06	11/30	35
0·04	11/29	36	0·04	8/30	25
0·02	7/30	21	0·02	2/30	4

* Correction made for mortality of 1/30 in control without pyrethrins

χ^2_2 for regression (not irradiated) = 1·33 ; $0{\cdot}7 > P > 0{\cdot}5$
χ^2_2 for regression (irradiated) = 1·03 ; $0{\cdot}7 > P > 0{\cdot}5$

To confirm these results, *E. cautella* adults were treated with series of irradiated solutions of pyrethrins and pyrethrins plus piperonyl butoxide (Table IV). Statistical analysis showed that there was no significant difference between these two sets of results. In other words, both series of solutions were degraded by the ultra-violet light to the same extent.

Conclusion and discussion

Under the experimental conditions described here, piperonyl butoxide does not prevent or reduce the loss of toxicity of pyrethrins brought about by irradiation with ultra-violet light.

Table III

Topical application to Ephestia cautella *adults of doses of* 0·05 *µl. of solutions of irradiated and non-irradiated pyrethrins plus piperonyl butoxide*

Concn. of pyrethrins, %	Concn. of piperonyl butoxide, %	Mortality Total	Mortality %*	Concn. of pyrethrins, %	Concn. of piperonyl butoxide, %	Mortality Total	Mortality %*
	Non-irradiated				Irradiated		
0·09	0·45	29/31	93	0·09	0·45	23/31	71
0·06	0·30	24/31	74	0·06	0·30	17/29	54
0·04	0·20	22/31	68	0·04	0·20	14/33	36
0·02	0·10	13/31	36	0·02	0·10	9/30	22

* Correction made for mortality of 3/29 in control with neither pyrethrins nor piperonyl butoxide

χ^2_2 for regression (not irradiated) = 1·05 ; $0·7 > P > 0·5$
χ^2_2 for regression (irradiated) = 1·86 ; $0·5 > P > 0·3$

Table IV

Topical application to Ephestia cautella *adults of doses of* 0·05 *µl. of irradiated solutions of pyrethrins and of pyrethrins plus piperonyl butoxide*

Concn. of pyrethrins, %	Concn. of piperonyl butoxide, %	Mortality Total	Mortality %	Concn. of pyrethrins, %	Concn. of piperonyl butoxide, %	Mortality Total	Mortality %
0·09	0	20/30	67	0·09	0·45	19/30	63
0·06	0	14/30	47	0·06	0·30	15/30	50
0·04	0	13/30	43	0·04	0·20	15/31	48
0·02	0	7/29	24	0·02	0·10	8/29	28

Control mortality with neither pyrethrins nor piperonyl butoxide, = 0/29

χ^2_2 for regression (pyrethrins only) = 2·66 ; $0·3 > P > 0·2$
χ^2_2 for regression (pyrethrins plus synergist) = 0·28 ; $0·9 > P > 0·8$
χ^2_6 for regression (combined results) = 1·27 ; $0·98 > P > 0·95$

For practical purposes it is important to distinguish between chemical change of the pyrethrins and any other loss of toxicity to insects brought about by exposure of the insecticide to ultra-violet light. The use of biological assay as described here is therefore particularly desirable because changes in the toxicity of the pyrethrins films were being directly examined.

If stabilisation occurs at all, it probably does so only to a very limited extent. In some instances of reported stabilisation, prolongation of the life of the film may, in fact, have been due to the increase in the ratio of piperonyl butoxide to pyrethrins which occurred as the pyrethrins were degraded. As the ratio increases, the film may retain its toxicity for a time to insects susceptible to pyrethrum synergists.

More detailed investigation of this point would require the refined use of methods of biological and chemical assay in conjunction.

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DETERMINATION OF MERCURY IN PLANT MATERIAL

By J. A. PICKARD and J. T. MARTIN

An analytical procedure used for the determination of traces of mercury in plant material is described. The conditions of combustion of organic matter are controlled to avoid loss of mercury and EDTA is used to reduce the values for apparent mercury on uncontaminated samples. Satisfactory recoveries of mercury added to apples, tomatoes and coffee are obtained.

Introduction

Organo-mercury compounds are used as sprays in this country to control disease of apple and pear and an aerosol form is employed under glass to protect the tomato plant. The low concentration of active ingredient used in the orchard spray leads to a low deposit on the fruits, which is reduced by subsequent weathering. Under glass, the application of aerosol is regulated, but the developing fruit may become contaminated. To assess whether treated crops are likely to offer a hazard to consumers, an analytical method is required that will determine up to 25 μg. of mercury in a large excess (50 g.) of plant material. For this sensitivity, the method must not only yield the mercury quantitatively, but must also show negligible values for apparent mercury on uncontaminated fruits.

The determination of traces of mercury in apples has been described by Abbott & Johnson,[1] who obtained recoveries of mercury of 90–94% and ' blank ' values of less than 0·5 μg. on 50-g. samples, both of which may be regarded as satisfactory. In connexion with work carried out on the control of the coffee berry disease, an examination was made of coffee beans taken from plants sprayed with a phenylmercury compound under experimental conditions only,[2] but difficulties arose with the method. The beans resisted oxidation in the period stipulated and combustion of the organic matter could only be completed by maintaining a high concentration of acid throughout the digestion process. The Abbott & Johnson procedure then gave an average recovery of 80% of mercury added to the beans, and the values on uncontaminated beans were unduly high (about 2 μg. per 50-g. sample). The reason for the incomplete recovery was therefore studied. The analytical procedure described below has given high recoveries of mercury from apple, tomato and coffee and negligible values for apparent mercury on uncontaminated samples.

Experimental

Destruction of organic matter

The combustion of the plant material is carried out by heating with a mixture of HNO_3 and H_2SO_4,[3] followed by treatment with hydrogen peroxide, with the incorporation of selenium throughout the reaction to fix the mercury.[4] Careful control of the conditions during the combustion process is necessary to avoid loss of mercury and digestion should be as complete as possible.[5] To overcome the difficulty caused by the water present in the plant material and the combustion products, which reduce the acid concentration and so may prevent the completion of the reaction, a trap is used to separate the water, as recommended by Klein.[6]

Laug & Nelson[3] pointed out that a violent reaction and excessive foaming in the early stage of the reaction may lead to loss of mercury and this has been our experience. Abbott & Johnson[1] showed that mercury may be carried away from the digestion mixture by a stream of carbon dioxide, but Gorsuch,[7] with different experimental conditions, found no such loss. Some workers[1, 3] suggest that the full heat of the bunsen burner should be used after frothing abates, but we have found that excessive heating may result in loss of mercury and prefer to give a more gentle treatment throughout the process, with an electrical heating mantle as a controlled source of heat.

If the material being destroyed reacts violently with the acid in the early stage of digestion, the acid mixture should be diluted with water. When samples froth excessively, the combustion should be started with HNO_3 sp. gr. 1·42 alone, the H_2SO_4 being added after partial digestion. Mercury may be lost if the fuming HNO_3 used in the later stage of digestion is added too rapidly

and vaporises too quickly from the simmering mixture. The slow addition of the fuming acid to a cooling digest is desirable, although this increases the time required for the completion of the reaction. Some materials are rapidly decomposed; others require prolonged treatment, but this is not conducive to loss provided it is gently carried out.

The use of hydrogen peroxide to complete the combustion has been discussed by Polley & Miller.[8] Perchloric acid should be avoided since volatile chloro-compounds of mercury may be formed and losses have also been attributed to the interaction of mercury with certain organic substances.[7] Some plants products resist oxidation in the final stage and fatty residues, some of which may be carried into the condenser, may occlude mercury and prevent its complete recovery.

Extraction of mercury from the digestion mixture

The mercury is extracted with dithizone under controlled conditions. This reagent is very sensitive but lacks specificity, as complexes are formed with many elements, and interference in mercury determinations has had to be overcome by careful control of pH or chemical treatment. In 1950, Vasak & Sedivec[9] showed that the only metal complexes of EDTA (disodium ethylenediaminetetra-acetate dihydrate) that will react with dithizone are those of univalent gold, silver, bivalent mercury and probably bivalent platinum. If present, gold and platinum after digestion would show higher valency states, so the problem becomes one of distinguishing between mercury and silver. If present as a contaminant, silver may be eliminated by treatment with potassium thiocyanate. Vasak & Sedivec pointed out that the incorporation of EDTA as a masking agent has an additional advantage in permitting the use of less pure reagents. We have used EDTA with success, extracting the mercury in the presence of the Complexone at pH 5.

Oxidation of the dithizone may occur during the extraction of the mercury immediately after the digestion process, but may be avoided by the use of sodium metabisulphite instead of hydroxylamine hydrochloride, but if sodium metabisulphite is used during the later dithizone extraction, incomplete recovery of the mercury may result.

Measurement of the mercury

The mono-colour method for the measurement of the mercury–dithizone complex is recommended by a Joint Committee of the Association of British Chemical Manufacturers and the Society for Analytical Chemistry[10] and is the method used by us. Although this Committee proposed a sodium hydroxide–hydroxylamine solution to remove excess of dithizone, we have continued to use ammonium hydroxide solution with satisfactory results.

The mercury–dithizone complex fades rapidly on exposure to bright sunlight, the colour partially returning if the solution is then placed in the dark. Care must therefore be taken to prevent exposure before the colour assessment and graduated flasks made of amber glass are used for the final solutions.

Preparation of reagents

Ammonium hydroxide solution (5%): Dilute aq. ammonia sp. gr. 0·880 (1 vol.) with water (19 vol.)

Buffer solution: Dilute 50 ml. of N-sodium acetate solution and 13 ml. of N-HCl to 250 ml.

Dithizone stock solution: Dissolve 50–60 mg. of dithizone in 20 ml. of carbon tetrachloride and filter. Extract with 100 ml. of 5% ammonium hydroxide solution and wash the alkaline layer twice with 5-ml. portions of carbon tetrachloride. Make the alkaline layer just acid with HCl, add 5 ml. of hydroxylamine hydrochloride solution and re-extract with 100 ml. of carbon tetrachloride. Wash the carbon tetrachloride solution twice with 15-ml. portions of water and store in the refrigerator.

For use, dilute ten times with carbon tetrachloride.

EDTA solution: 4%.

Hydroxylamine hydrochloride solution: 20% w/v. Extract with small quantities of diluted dithizone solution until the extract is colourless after the removal of excess dithizone with 5% ammonium hydroxide solution.

Sodium metabisulphite solution: 20% w/v. Treat as hydroxylamine solution. The extraction is slow and a mechanical shaker is helpful. Filter the bisulphite solution through a wetted Whatman No. 54 paper to remove entrained carbon tetrachloride.

Sodium hypochlorite solution: Titrate the approx 15% commercially available product, and dilute to 5% available chlorine. Store in refrigerator.

Sodium thiosulphate solution: 1·5%, freshly prepared.

Apparatus

A 500-ml. two-necked round-bottomed flask carrying a 50-ml. cylindrical separating funnel by a B.19 joint in the side neck and a 250-ml. Soxhlet solvent trap (Loughborough Glass Co. Ltd. Catalogue No. ESR/25/55) by a B.34 joint in the central neck. The solvent trap supports a 8–10 × 1 in. air condenser, to the top of which is fitted a double-surface water condenser by a B.24 joint.

A 500-ml. 250-W heating mantle, controlled by an auto-transformer.

Procedure

To 25–50 g. of a representative sample in the reaction flask add 0·1 g. of selenium powder and a few glass beads. Add a mixture of 10 ml. of H_2SO_4 sp. gr. 1·84 and 10 ml. of HNO_3 sp. gr. 1·42, and digest slowly at low heat with the tap of the solvent trap open. Carefully add more HNO_3 sp. gr. 1·42 if there is any indication of charring. As soon as the initial reaction abates, close the tap and continue the digestion with fuming HNO_3 with sufficient heat to ensure simmering and maintaining oxidising conditions until the organic matter is destroyed and the solution is clear.

Allow the digest to cool and run the acid solution in the trap into the flask. Add 20 ml. of 30% w/v hydrogen peroxide and reflux gently for 30 min. Wash down the condenser, trap and separating funnel with 50 ml. of water, cool to room temperature and filter through a Whatman No. 541 filter paper. Adjust the filtrate to 200 ml. with water.

Transfer a suitable aliquot (100 ml. or less according to the level of mercury expected) to a 400-ml. beaker and just neutralise to litmus with ammonia solution, sp. gr. 0·880, with cooling in an ice bath. Make just acid to litmus by the dropwise addition of HCl and add 10 ml. of sodium metabisulphite solution, 25 ml. of buffer solution and 5 ml. of EDTA solution. The pH of the solution should be 5.

Transfer to a 250-ml. separating funnel, adjust the volume to about 200 ml. with water and extract with 10-, 5- and 5-ml. portions of diluted dithizone solution. Transfer the dithizone extracts to a 100-ml. separating funnel containing 50 ml. of 0·1N-HCl. Add 2 ml. of sodium thiosulphate solution and shake for 1 min. Run off the carbon tetrachloride layer and discard. Wash the acid layer with two 3-ml. portions of carbon tetrachloride and discard the washings. Add 3 ml. of sodium hypochlorite solution and shake for 1 min. Add 5 ml. of hydroxylamine hydrochloride solution, shake vigorously for 1 min., blow off any chlorine and shake again. Extract twice with 3-ml. portions of carbon tetrachloride and reject the extracts.

Add 20 ml. of buffer solution, 2 ml. of EDTA solution and mix. Add 7 ml. of carbon tetrachloride and 1 ml. of diluted dithizone solution and extract with vigorous shaking. If necessary add diluted dithizone solution a few drops at a time until an excess of dithizone is present. Repeat the extraction using 4 ml. of carbon tetrachloride and a few drops of dithizone solution.

Combine the dithizone extracts in a 60-ml. separating funnel and extract the excess of dithizone by shaking twice with 3 ml. portions of 5% ammonium hydroxide solution. Run the carbon tetrachloride solution containing the mercury–dithizone complex through 1–2 g. of anhydrous sodium sulphate into a 20-ml. graduated flask made of low-actinic amber glass and adjust the volume to 20 ml. with carbon tetrachloride. Measure the optical density at 490 mμ.

Calibration with mercuric chloride

Known amounts of mercury, as mercuric chloride in aqueous solution, were added to 50-ml. portions of 0·1N-HCl. Hydroxylamine hydrochloride solution (5 ml.), buffer solution (20 ml.) and EDTA solution (2 ml.) were added, the mercury was extracted with dithizone and the analysis continued as described. The optical densities recorded in a 2-cm. cell are as follows:

Mercury, μg.	0	5	10	15	20	25
Optical density	0·004	0·175	0·343	0·516	0·685	0·864

Recoveries of mercury from food samples

Mercury was added, as mercuric chloride, to apple fruits, peel or flesh (50-g. samples), tomato fruits (50-g. samples) and roasted, ground coffee (25-g. samples) before digestion. The recoveries, after allowing for control values, are given in Table I.

Table I

Recovery of mercury from apple, tomato and coffee

Mercury added, μg.	Mercury found, μg.				
	Apple fruit	Apple peel	Apple flesh	Tomato	Coffee
0	0·1	—	—	0·6	0·5
	0·2	—	—	0·7	0·75
	0·3	—	—	—	—
5	5·2	5·0	4·9	5·0	4·8
				5·1	4·8
10	10·2	10·1	10·1	9·9	9·4
	10·0			10·0	9·8
15	14·9	15·0	15·0	14·9	14·4
	15·0			14·9	14·5
20	20·1	20·0	20·0	19·7	19·3
	20·2			20·1	19·5
25	—	25·1	25·0	—	—

Conclusion

The recoveries in all the tests, which covered a range up to 0·5 p.p.m. on 50 g. samples, were satisfactory. The mean value recorded for apparent mercury on uncontaminated apple fruits was equivalent to 0·004 p.p.m., on tomatoes 0·01 p.p.m. and on ground coffee (25 g. samples) 0·025 p.p.m.

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CORRELATIONS BETWEEN THE pH VALUE OF MEAT AND THE DIFFUSION OF SALT

By L. KÖRMENDY and Gy. GANTNER

A study is made of the speed of penetration of salt into loin pork meat under constant conditions of pickling (weight and size of sample, ratio of pickle to meat, temperature), and the results statistically analysed. Under the conditions employed, the salt content of the meat after 24 h. is considered to be characteristic of the penetration rate of the salt.

There was no close correlation between the pH of the meat and the 24-h. salt content, but the pH values fell within a rather narrow range (87% between 5·4 and 6·7).

Introduction

Bate-Smith[1] and Callow[2] made interesting observations on the correlations between the pH value of meat and its electrical resistance. Amongst other facts they found that a high electrical resistance in meat is associated with a high pH value. This is attributed to the fact that an increase in pH results in swelling of the muscle fibres, as a consequence of which the spaces between cells and fibres, through which ions can freely migrate, contract. Callow distinguishes ' open ' structures of compact consistency on the one hand and ' stringy ', ' flaccid ', ' closed ' structures, which are dry to the touch, on the other. According to Callow & Ingram[3] these phenomena are also correlated with the ability of the meat to be cured. Meat of lower pH value and lower electrical resistance is more easily permeable to brine. From this it naturally follows that it is advisable to select, treat and store the raw materials used for meat curing so that they have an acid pH, as such raw material is more suitable for curing.

The results of Gibbons & Rose[4] deserve special mention : starting from raw materials of lower pH values they found a somewhat more favourable development of colour on pickling ; they obtained, however, no close correlation between the pH value and the salt content. It must be remembered, however, that as mentioned in their paper, their test conditions were not constant (e.g., the samples tested differed from each other in thickness etc.).

In a previous paper[5] were reported the results of experiments on the behaviour of fresh, warm, raw meat of high pH value on curing. It was established that generally there is no sensible difference between fresh raw meat, 1·5–2 h. after slaughtering and meat stored for 24 h. or longer, in respect of change of salt penetration with time. This was so in spite of the fact that essential differences were found as regards pH value, texture, etc. Also to be taken into account was the fact that fresh beef of high pH value has to be considered from a biochemical angle as being in a ' labile ' condition. Biochemical changes (decomposition of glycogen and ATP, formation of lactic acid, etc.) set in very rapidly during the first 24 h. after slaughtering, but during subsequent cold storage, changes in the physico-chemical properties, consistency and pH value of the meat are considerably slower. It is therefore obvious that during early curing, particularly within the first 24 h., such changes take place simultaneously with salt penetration. We do not know, however, in what way these processes are influenced by the penetration of salt into the tissues.

In this paper experiments are described in which was studied the correlation between ease of curing and pH for specimens of meat which had been kept for at least 48 h., after which their pH value changed at a comparatively slow rate. Such samples were, therefore, biochemically relatively more ' stable '.

Experimental

Materials and methods

In all cases loin pork was used so as to ensure uniformity of test material. It was kept for 2–6 days at +2 to +6° c and was trimmed free of fat and coarse connective tissue was removed before being sampled. Cylindrical samples of 10 cm.-length were cut by means of a steel tube with sharpened end, of 5 cm. diameter, so that the diameter of the samples was relatively uniform. The samples were stored at +2° to +5° c until required.

As shown in Fig. 1, in spite of the precautions taken, the samples were of different weight (i.e., of different diameters) according to the quality of the meat, the lengths being practically identical.

The weight of the 10-cm. cylinders (102 pieces) varied between 122 and 200 g. (mean value 158 g.), but the standard deviation, $S = \pm 14{\cdot}4$ (9·1%) was not very excessive. In Fig. 1 the probit analysis indicates an approximately normal distribution. As the curing time necessary for reaching a given average concentration of salt is roughly proportional to the square of the diameter, i.e., in this case to the weight of the cylinders, an attempt was made to eliminate the effect of weight fluctuations on the speed of the curing process by calculating the partial correlation coefficients.[6]

In practice, a 1 : 1 ratio of pickle to meat is used and this was followed in all the tests made. The pickle $20{\cdot}4 \pm 0{\cdot}2$ g. of NaCl and $0{\cdot}4 \pm 0{\cdot}03$ g. of KNO_3 per 100 ml.

The samples were placed in 400-ml. beakers and the necessary volume of pickle (cooled to 2–5°) was added from a graduated cylinder. The beakers were covered with watchglasses and kept for 24 h. at 2–5° C, the pickle not being disturbed during this time. After this storage, the concentration of salt was determined by use of the equation

$$C_{24} = p(d_0 - d_{24})$$

where C_{24} = average salt concentration of meat (g./100 ml.) after 24 h.
p = ratio of ml. of pickle/ml. of meat
d_0 = initial salt concentration of pickle (in these tests 20·4 g./100 ml.)
d_{24} = salt concentration of pickle after 24 h. (g./100 ml.)

The value p is given by

$$p = p' . S_0$$

where p' = ml. of pickle/g. of meat (=1·0 in these tests)
S_0 = average specific gravity of salt-free meat (1·067 in reference [7])

The above equation is only strictly valid if no change of volume occurs during the process of pickling. Actually it was found that the meat swelled 3–4% after curing for 24 h. and in view of the fact that identical conditions were used in all tests, the error caused by this was disregarded.

The optimum time for determining the salt contents was particularly considered. It may be demonstrated by a calculation of errors[7] that, with diffusion processes the value of the diffusion constant can be determined with minimum error at the mid-point of the process; in the case of prolonged diffusion times, determinations will become inaccurate in the vicinity of the equilibrium concentration. As shown in Fig. 2 the salt concentrations found after 24 h. with the 10-cm. sample pieces are situated rather far from the equilibrium concentration.

Hence, for comparative work, the salt concentration reached after 24 h. (C_{24}) could be considered as giving a good approximate measure of the speed of diffusion.

The pH values of the raw meat free of salt were determined with a quinhydrone electrode. The change of the pH value during the curing process was not followed, nor was any particular attention given to the development of colour.

Results and conclusions

In all, 102 sample pieces of different quality were tested and the results are shown in Figs. 3 and 4. The following conclusions may be drawn from the results:

(1) There is no close correlation between the pH value of the meat and speed of salt penetration (C_{24})—neglecting changes in diameter of the samples. The value of the correlation coefficient between pH and C_{24} (0·276) is very low, but it appeared highly significant.

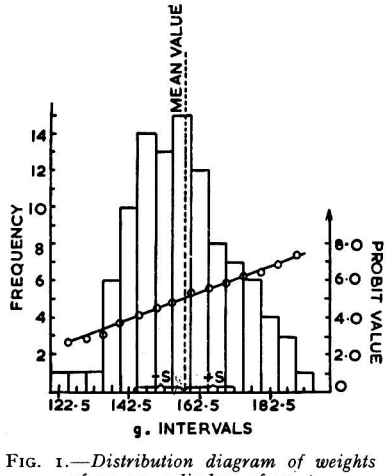

FIG. 1.—*Distribution diagram of weights of 10-cm. cylinders of meat*
S = Standard deviation

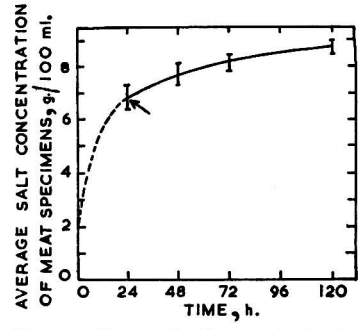

FIG. 2.—*Change of salt concentration in cylinders of meat as function of the time of curing*
No. of samples tested 11; weight of cylinders 125–180 g. (average 152 g.); length of cylinders 10 cm.

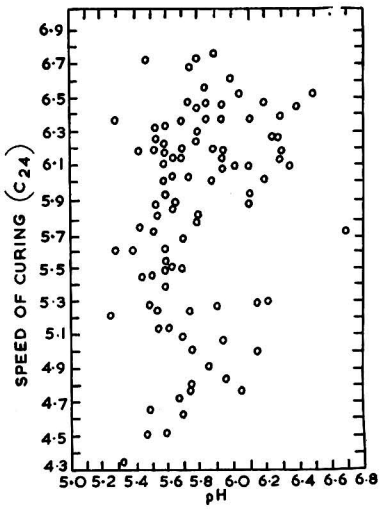

FIG. 3.—*Relation between the pH value of meat and the speed of curing* (C_{24})
$r = 0{\cdot}276$

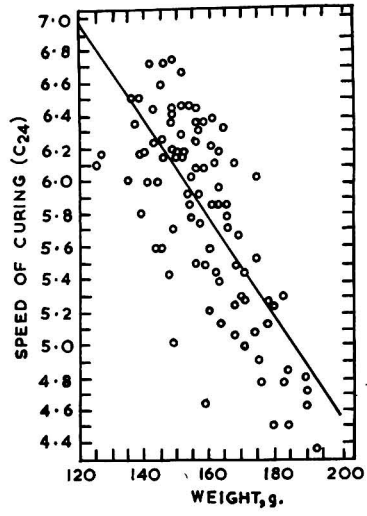

FIG. 4.—*Relation between the weight (g.) and the speed of curing* (C_{24}) *of the meat showing regression line*
$r = 0{\cdot}74$ $C_{24} = 0{\cdot}03 \times$ weight (g.) $+ 10{\cdot}57$

(2) A rather close correlation exists between the speed of salt penetration (C_{24}) and the weight (g.) of the cylinders which is proportional to the square of the diameters.

The value of the correlation coefficient between weight in g. and C_{24} ($-0{\cdot}74$) is relatively high and very highly significant.

(3) Even eliminating the fluctuations of diameter (i.e., assuming constant diameter), there is no correlation between the pH value of the meat and the speed of salt penetration. The partial correlation coefficient for C_{24}, pH and weight (0·19) is not significant.

(4) Eliminating the fluctuations between pH values (for samples of identical pH value) there is a very highly significant, close correlation between the speed of salt permeation and the weight of the pieces of meat (r for C_{24}, weight and pH = 0·727).

There is probably no linear connexion between the C_{24} and weight values. The cylinder diameters having, however, fluctuated within not too wide limits (see Fig. 1) in the range tested, the linearity test did not indicate this.

Under the experiment conditions described, it was not possible to demonstrate a close, statistically supported correlation between the pH values of the meat specimens tested and the permeability to salt solutions of the muscle tissue. It should be mentioned that the samples tested had pH within a relatively narrow range (87% between pH 5·4 and 6·3 and none between 6·8 and 7·0). In view of the small pH changes, it is possible that the meat specimens did not show sufficiently large differences in their structural characteristics, and for that reason no differences in the speed of the diffusion of salt could be demonstrated.

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USE OF GAS CHROMATOGRAPHY IN MEASURING THE ETHYLENE PRODUCTION OF STORED APPLES

By D. F. MEIGH

A method has been developed, using a gas chromatograph with a flame ionisation detector, for estimating ethylene in 0·5-ml. samples of air at concentrations down to 1 p.p.m. It has been used to follow the ethylene production of apples stored under various conditions.

Introduction

Although it has long been known that ethylene can be used to stimulate the ripening of fruits and that during their respiratory climacteric many fruits themselves produce ethylene, the problem whether ethylene, under natural conditions, plays an active rôle in initiating the ripening process has not been solved. This may largely be due to the difficulty of estimating the small quantities of ethylene that are produced. The principal methods that have been used are (*a*) bromination,[1] (*b*) wet oxidation[2] and (*c*) absorption in mercuric perchlorate followed by release of the ethylene with HCl and manometric measurement of the gas.[3] For most experiments with ripening fruits these methods require, for a reliable estimation, the accumulation of ethylene from several litres of gas.

The advent of gas chromatography has made a new technique available. In some preliminary investigations of this method a katharometer was tried as detector, but it was found insufficiently sensitive for quick routine estimation. The recent development of more sensitive detectors[4–8] has enabled this disadvantage to be overcome. It should be noted however that while this work was in progress Burg & Thimann[9] have been able, by a number of refinements, to obtain satisfactory results with a katharometer detector.

Experimental

Apparatus

Two detectors were used, one fitted to the analytical column and the other to a balancing column.

The column packing consisted of Johns–Manville C22 firebrick, ground and sieved. The 36–60 mesh range was retained and impregnated with 30% of its weight of liquid paraffin as the stationary phase. The analytical column consisted of eight 3-ft. (92 cm.) lengths of 5-mm. bore glass tubing joined end to end with 1-mm. bore capillary **U**-tubes. The balancing column, which was used to offset the appreciable background caused by the slow evaporation of the stationary phase, and to cancel out any slow alterations in the eluent gas composition, consisted of a **U**-shaped 6 ft. length of 2-mm. bore capillary tubing packed with sufficient of the firebrick to give it the same resistance to gas flow as the analytical column. The columns were suspended in an unheated jacket and the whole apparatus housed in a constant temperature room at 20°.

Two flame ionisation detectors were made (Fig. 1). Tinned cans were used as containers and the general design of McWilliam & Dewar[8] followed. Air for ventilating the containers was supplied by an Austen diaphragm pump and filtered. A flow rate of about 15 l./h. for each detector was sufficient to prevent condensation of water vapour on the metal parts. The positive potential of 200V for the needles was supplied from the power pack of the amplifier and the outputs from the detector gauzes were fed through 18-in. lengths of coaxial cable to the 10,000 megohm input resistors of a current amplifier, which was constructed to the design of Lovelock.[6] A similar circuit has been described by Thompson,[7] who gives useful advice on the construction of this type of amplifier. The most stable conditions were secured by taking the 300-V d.c. supply from a Solartron power supply unit type AS-516 and obtaining the current for the valve filaments from a Servomex voltage stabiliser type D.C. 65. The output from the amplifier was recorded on a 3-mV potentiometric recorder with an 11 in. wide chart. To provide recorded peaks of convenient height over a wide range of ethylene concentrations, the amplifier output was varied by means of a voltage attenuator.

The eluent gas consisted of approximately equal parts by volume of nitrogen and hydrogen obtained from cylinders, the supply pressure being controlled at each cylinder by a reducing

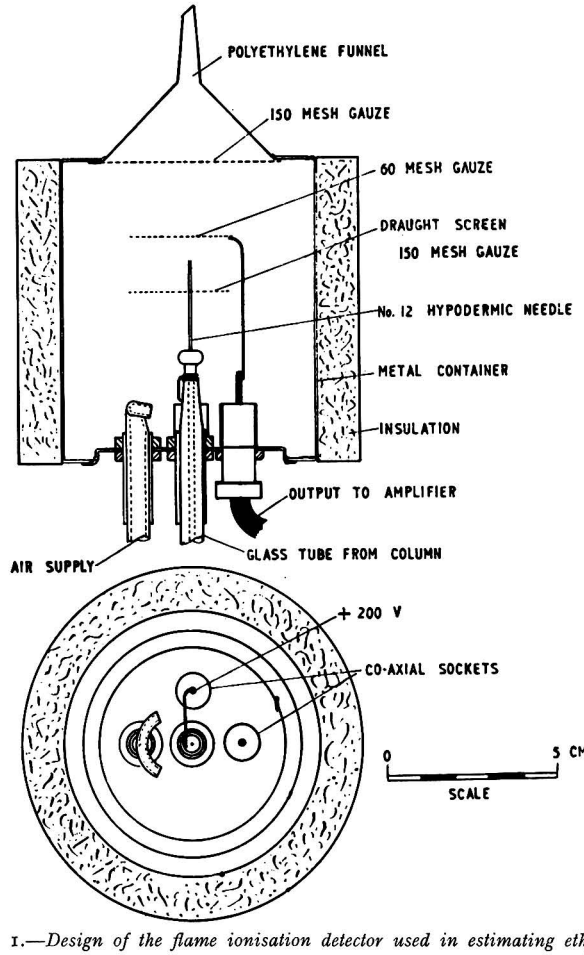

FIG. 1.—*Design of the flame ionisation detector used in estimating ethylene*

valve and an Edwards VPC1 pressure controller. The flow rate of each gas was measured before mixing by passage through an orifice flowmeter and the pressure of the mixed gases was measured on a mercury manometer. The gas mixture then passed through a rotameter flowmeter ahead of each column.

Analysis of air samples

It was found that an alteration in the composition of the eluent gas affected the response of the detectors, an increase in the proportion of hydrogen increasing the response. Therefore for quantitative analyses the pressure controllers were adjusted to give a standard flowrate ratio and a column inlet pressure of 29·8 cm. Hg.

The chosen column was of such a length that under the operating conditions air, carbon dioxide and other hydrocarbons were well separated from ethylene. The response of the detector to air and carbon dioxide, which was to produce a negative peak on the recorder, was only apparent at high sensitivity settings. Retention times for the compounds which are most difficult to separate from ethylene were air 5, methane 6, carbon dioxide 7, acetylene 7·5, ethylene 9, ethane 11, propylene 20, propane 22 min. Where only air, carbon dioxide and ethylene were present in appreciable quantities in the samples it was found possible to double the speed of analysis by admitting one sample every 5 min. without causing any interference between peaks. The recorder chart reproduced in Fig. 2 shows four analyses in which this overlapping method was used.

The device for introducing gas samples (Fig. 3) had a volume of about 0·5 ml. If the air samples were much larger than this the emergence of the air fraction extinguished the detector flame. Air samples were admitted to the device from glass gas-sample tubes by displacement with mercury.

The apparatus was calibrated with ethylene–air mixtures of known composition. From the curve in Fig. 4, which relates amplifier response (i.e., peak height) to ethylene concentration, it can be seen that the response of the detector is virtually linear from 1 to 1000 p.p.m. Instability of the base line made it impracticable to analyse mixtures containing less than 1 p.p.m. of ethylene.

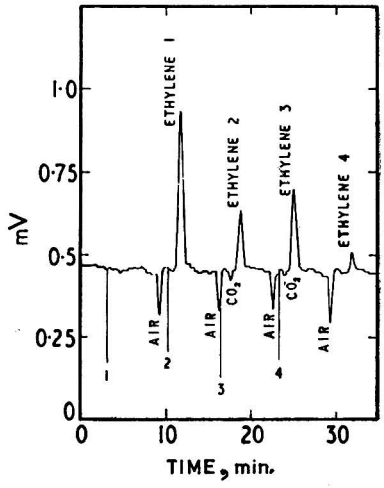

FIG. 2.—*Chart showing the analysis of four ethylene–air mixtures*
(1) 17 p.p.m. (2) 7 p.p.m. (3) 9 p.p.m. (4) 2 p.p.m. of ethylene

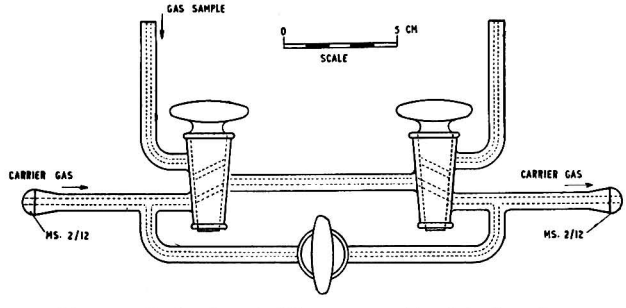

FIG. 3.—*Device for admitting gas samples into the column*

Application to the study of stored apples

The use of the method can be illustrated by some results obtained during the 1958–9 storage season with samples of air from five apple stores.

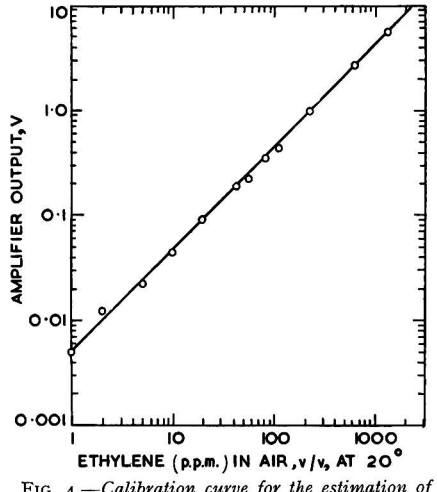

FIG. 4.—*Calibration curve for the estimation of ethylene in air*

Edward VII apples picked at the normal date from an orchard at Sutton Valence were sorted, the unsound fruit removed, and the contents of each orchard box equally distributed between the boxes for each experiment. The fruit was stored on a half-ton scale in steel gas-tight cabinets at 3°, according to methods described elsewhere.[10] By appropriate ventilation with air the carbon dioxide concentrations in the five cabinets were adjusted to the following values (1) 0·5%, (2) 4%, (3)–(5) 8–9%. The apples for Cabinet 4 were kept at room temperature for one week before storage, while those for Cabinet 5 were enclosed in an airtight box at room temperature for one week before storage and ventilated during that time at a rate of 500 l./h. with air containing 500 p.p.m. of ethylene.

From the rate of air ventilation through each cabinet, recorded daily, and the concentration

of ethylene in the air samples, estimated weekly, the rate of production of ethylene by the fruit was calculated. After removal from store half the apples from each cabinet were examined for superficial scald, the other half being kept at room temperature for 3 weeks for a second examination. The results of these are shown in Table I. The apples in Cabinets 1 and 2 were stored for shorter periods of time than the rest so that when unloaded they should be of comparable maturity. The variation in scald incidence between the different treatments is typical. The apparent decrease in scald in the fruit of Cabinet 5 between first and second examinations is, however, probably due to the obliteration of the skin by heavy rotting. In Table II are shown the figures for estimated concentration of ethylene in three of the cabinets and in Fig. 5 are shown the rates of production of ethylene in all five cabinets. These values are of the same order of magnitude as those obtained for cold-stored apples by Fidler[11] and Gerhardt.[12]

Table I

Incidence of scald in apples stored at 3° under various conditions

Cabinet	Treatment	Storage date	Days in store	No. of apples affected by scald, %	
				1st examination	2nd examination
1	0·5% CO_2	5 Nov. 58	139	38·9	55·7
2	4% CO_2	,,	169	16·0	27·2
3	8–9% CO_2	,,	201	1·15	1·2
4	8–9% CO_2 after keeping 1 week at room temp.	13 Nov. 58	193	16·15	16·1
5	8–9% CO_2 after keeping 1 week at room temp. with ethylene ventilation	,,	193	27·1	18·6

Table II

Concentration of ethylene in stores of Edward VII apples, 1958–9

Days in store	Cabinet no. 1	2	3	Days in store	Cabinet no. 1	2	3
2	10		102	93	29	121	212
9	—	80	126	100	24	90	149
16	23	103	129	107	21	86	—
23	29	155	182	114	23	96	140
30	29	—	226	121	21	83	121
37	31	169	209	128	27	84	143
44	29	140	179	135	21	94	130
58	34	137	165	149	—	77	102
65	35	161	—	156	—	77	102
72	32	137	217	163	—	81	55
79	32	134	204	170	—	—	86
86	35	130	209	177	—	—	110

The results for Cabinets 1, 2 and 3 show the effect of carbon dioxide concentration on the rate of production of ethylene. Thus the ratio for 0·5% : 4% : 8% carbon dioxide is about 10 : 4 : 2·5. The corresponding ratio for carbon dioxide production during the major period of storage after the rate had levelled off was 10 : 7·5 : 6·5. This marked difference does not seem to be connected with the reduced partial pressure of oxygen which also results from the method of storage. Burg & Thimann[9] have shown that in the apple, ethylene production and oxygen consumption are affected in an almost identical manner by alterations in the partial pressure of oxygen. Since Fidler has shown[11] that Edward VII apples stored in 8% carbon dioxide at 38° F have a respiratory quotient of 1·0–1·1 during the major part of storage it can be concluded that the high carbon dioxide concentration is responsible for the greater retarding effect on ethylene production than on carbon dioxide production.

A comparison of the three lots of apples stored in 8–9% carbon dioxide shows that during the first 80 days of storage the apples kept at room temperature before storage produced much more ethylene than those stored immediately. Treatment with ethylene before storage, however, did not have any further stimulating effect, presumably because the delay alone was quite sufficient to initiate the climacteric rise in respiration rate. It will be seen from the figures for incidence of scald given in Table I that there is some correlation between ethylene production and incidence of scald, but this may not have any significance.

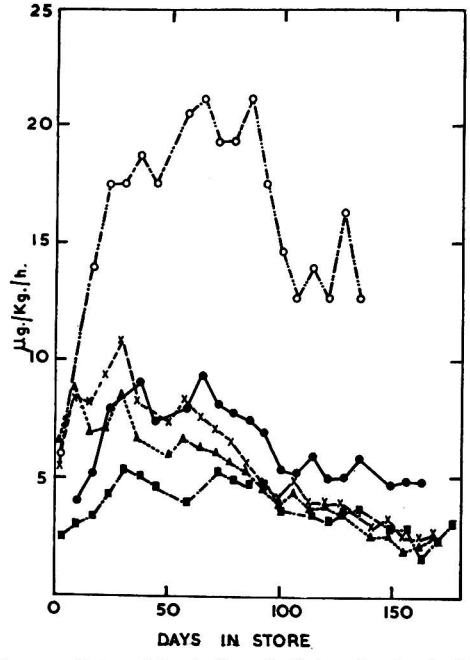

FIG. 5.—*Rates of production of ethylene by stored apples*

○- · · —○ Cabinet 1 0·5% CO_2
●——● Cabinet 2 4% CO_2
■— · —■ Cabinet 3 8–9% CO_2
× — — × Cabinet 4 8–9% CO_2, kept 1 week at room temperature before storage
▲ — — ▲ Cabinet 5 8–9% CO_2, kept 1 week at room temperature with ethylene treatment before storage

Summary

(1) Gas chromatography has been used to analyse the ethylene produced by stored apples. This was achieved by using a sensitive flame ionisation detector and a column long enough to separate ethylene from possible interfering substances.

(2) Measurements of the rate of ethylene production by apples stored in various concentrations of carbon dioxide show that high carbon dioxide concentrations have a greater retarding effect on ethylene production than on carbon dioxide production.

(3) Apples kept at room temperature for a week before storage produced ethylene at an abnormally high rate for about the next 80 days.

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GIBBERELLIC ACID. XI.*—The Growth-Promoting Activities of Some Functional Derivatives of Gibberellic Acid

By J. S. MOFFATT and MARGARET RADLEY

A number of derivatives of gibberellic acid have been prepared and their growth-promoting activities compared. Eleven salts and four acyl derivatives showed activities similar to that of gibberellic acid when applied to the roots or to the leaves of dwarf pea seedlings. Five esters and nine acyl derivatives of esters showed no activity when applied to the leaves; most of them were moderately active when applied, in aqueous culture solutions, to the roots possibly because they underwent slow hydrolysis.

Introduction

In view of the marked growth-promoting effects of gibberellic acid it was considered to be of interest to prepare and examine a series of derivatives for similar properties. Although Takahashi *et al.*[1] considered that the methyl esters of three gibberellins were inactive, Bukovac & Wittwer[2] found that methyl gibberellate and methyl gibberellin A_1 were active but much less so than their parent acids. Some of the results given below have already been mentioned in patent specifications.[3]

Experimental

Preparation of derivatives of gibberellic acid

Ammonium gibberellate monohydrate was obtained as a colourless powder, m.p. 167–170° (decomp.) on evaporation under reduced pressure at 20–24° of a solution of gibberellic acid (100 mg.) in 0·135N-ammonium hydroxide (3·25 ml.) (Found: on a sample dried at 80°/12 mm.: C, 59·8; H, 7·0. $C_{19}H_{25}O_6N,H_2O$ requires C, 59·8; H, 7·1%).

Sodium gibberellate sesquihydrate, m.p. 232–238° (decomp.) (Found on a sample dried over P_2O_5 in a vacuum desiccator: C, 57·4; H, 6·15. $2C_{19}H_{21}O_6Na,3H_2O$ requires C, 57·7; H, 6·1%) and *potassium gibberellate sesquihydrate*, m.p. 225–229° (decomp.) (Found: C, 55·6; H, 5·7. $2C_{19}H_{21}O_6K,3H_2O$ requires C, 55·5; H, 5·9%), which formed hygroscopic powders were obtained on neutralisation of solutions of gibberellic acid (100 mg.) in methanol (3 ml.) with the appropriate 0·02N alkali hydroxide followed by evaporation of the solutions at 18–20° under reduced pressure.

Rubidium gibberellate. A solution of gibberellic acid (87·7 mg.), in methanol (1 ml.), was treated with a solution of rubidium carbonate (26·6 mg.) in water (0·2 ml.) and the mixture filtered. The filtrate was evaporated at room temperature under reduced pressure and the residue repeatedly extracted with warm acetone. The residual *salt* had m.p. 213–216° with previous decomp. (Found: C, 53·1; H, 5·5. $C_{19}H_{21}O_6Rb$ requires C, 53·0; H, 5·0%).

The following salts were prepared by shaking for periods of 5–15 h. suspensions of gibberellic acid (100 mg.) in water (5 ml.), with a large excess of the appropriate metallic carbonate. The mixtures were then filtered and the salts obtained on evaporation of the filtrates at room temperature under reduced pressure. The salts were dried for several days in a vacuum desiccator before analysis.

Copper gibberellate trihydrate consisted of a blue-green solid, m.p. 245–248° (decomp.) [Found: C, 56·0; H, 5·7. $(C_{19}H_{21}O_6)_2Cu,3H_2O$ requires C, 56·4; H. 6·0%]. *Silver gibberellate dihydrate* formed colourless needles (from water), m.p. 198–199° (decomp.), which turned brown on exposure to light (Found: C, 46·7; H, 5·1; Ag, 21·7. $C_{19}H_{21}O_6Ag,2H_2O$ requires C, 46·6; H, 5·2; Ag, 22·1%). *Calcium gibberellate trihydrate* was a solid, m.p. 260° [Found: C, 58·7; H, 6·0. $(C_{19}H_{21}O_6)_2Ca,3H_2O$ requires C, 58·2; H, 6·2%]. *Lead gibberellate monohydrate* had m.p. 215–218° (decomp.) [Found: C, 49·8; H, 5·0. $(C_{19}H_{21}O_6)_2Pb,H_2O$ requires C, 49·8; H, 4·8%]. *Manganese gibberellate trihydrate* formed a pale-fawn solid, m.p. 261–266° (decomp.) [Found: C, 57·5; H, 5·8. $(C_{19}H_{21}O_6)_2Mn,3H_2O$ requires C, 57·1; H, 6·0%]. *Cobalt gibberellate trihydrate* consisted of a purple solid, m.p. 204–206° (decomp.) [Found: C, 56·8; H, 5·7. $(C_{19}H_{21}O_6)_2Co,3H_2O$ requires C, 56·8; H, 6·0%]. *Gibberellic acid cyclohexylamine salt* (Cross[4])

* Part X: Morrison, A., & Mulholland, T. P. C., *J. chem. Soc.*, 1958, p. 2702

separated out in felted needles, m.p. 173–177° (decomp.), on treatment of gibberellic acid (106 mg.) in acetone with cyclohexylamine (34 mg.) in acetone. Consistent analysis results were not obtained for this compound. It gave back gibberellic acid on acidification with dilute HCl.

Esters of gibberellic acid

Methyl gibberellate has been previously described.[5] The following homologues were prepared by the interaction of gibberellic acid, in methanol, and the appropriate diazohydrocarbon in ether.

Ethyl gibberellate formed prismatic needles (from methanol–ether), m.p. 150–151°, of the hemi-methanolate (Found, on a sample dried at 100°/12 mm.: C, 66·6; H, 7·6. $C_{21}H_{26}O_6,\frac{1}{2}CH_4O$ requires C, 66·2; H, 7·2%), which on being further dried at 140°/12 mm. afforded the solvent-free ester, m.p. 155–156° (Found: C, 67·3; H, 7·2. $C_{21}H_{26}O_6$ requires C, 67·4; H, 7·0%). *Isopropyl gibberellate* formed needles (from methanol–ether), m.p. 151–152°, of the hemimethanolate (Found, on a sample dried at 100°/12 mm.: C, 66·5; H, 7·8. $C_{22}H_{28}O_6,\frac{1}{2}CH_4O$ requires C, 66·8; H, 7·5%). *Butyl gibberellate* formed needles (from methanol–ether), m.p. 153–156°, of the hemimethanolate (Found, on a sample dried at 100°/12 mm.: C, 67·3; H, 7·7. $C_{23}H_{30}O_6,\frac{1}{2}CH_4O$ requires C, 67·4; H, 7·7%). *Octyl gibberellate* was prepared from gibberellic acid and diazo-octane in isopropyl ether and formed prismatic needles (from ether–light petroleum, b.p. 40–60°), m.p. 157–158° (Found: C, 70·2; H, 8·3. $C_{27}H_{38}O_6$ requires C, 70·7; H, 8·35%).

Acyl derivatives of gibberellic acid

Acetylgibberellic acid has been previously described.[5] *Butyrylgibberellic acid.* Gibberellic acid (50 mg.), in dry pyridine (0·5 ml.), was treated with butyric anhydride (0·1 ml.) and the mixture stored for 23 h. It was treated with methanol (0·5 ml.) and then evaporated at 25° under reduced pressure and the residual gum extracted with 2N-potassium hydrogen carbonate (1·5 ml.). Acidification of the extract with dilute HCl yielded a precipitate which was collected, washed with water and then crystallised from ethyl acetate–light petroleum (b.p. 40–60°) to give the *acyl derivative* as rhombs (35 mg.), m.p. 192° (Found, on a sample dried at 140°/12 mm.: C, 66·1; H, 7·0. $C_{23}H_{28}O_7$ requires C, 66·3; H, 6·8%). *Benzoylgibberellic acid* was similarly prepared with benzoyl chloride in pyridine. It formed needles (from methanol–ether), m.p. 204–205° (Found: C, 69·2; H, 5·9. $C_{26}H_{26}O_7$ requires C, 69·3; H, 5·8%). *Diacetylgibberellic acid.*[4] A solution of acetylgibberellic acid (35 mg.) in dry pyridine (0·55 ml.) and acetic anhydride (0·35 ml.) was kept for 5 days and then evaporated at 90° under reduced pressure. The residue was extracted with sodium bicarbonate solution. Acidification of the extract with dilute HCl yielded a solid (28 mg.; m.p. 160–168°), which, on repeated recrystallisation from ether–light petroleum, afforded the diacetyl derivative as prisms, m.p. 186–187° (Found: C, 64·3; H, 6·2. $C_{23}H_{26}O_8$ requires C, 64·2; H, 6·1%).

Acyl derivatives of esters of gibberellic acid

Methyl acetylgibberellate has been previously described.[5] *Ethyl acetylgibberellate,* prepared by treatment of acetylgibberellate in methanol with ethereal diazoethane, formed rhombs (from methanol–ether), m.p. 203° (Found: C, 66·0; H, 6·85. $C_{23}H_{28}O_7$ requires C, 66·3; H, 6·8%). *Isopropyl acetylgibberellate,* prepared with ethereal diazo-isopropane, formed prisms (from ether), m.p. 202–203° (Found: C, 66·7; H, 7·05. $C_{24}H_{30}O_7$ requires C, 67·0; H, 7·0%). *Butyl acetylgibberellate,* prepared with ethereal diazobutane, formed prisms (from ethyl acetate–light petroleum b.p. 40–60°), m.p. 147–149° (Found: C, 67·1; H, 7·2. $C_{25}H_{32}O_7$ requires C, 67·5; H, 7·3%). *Octyl acetylgibberellate* was prepared by the action of acetic anhydride and pyridine on octyl gibberellate, and consisted of a viscous oil, b.p. 195–202° (bath temperature)/5×10^{-5} mm. (Found: C, 69·6; H, 8·0. $C_{29}H_{40}O_7$ requires C, 69·6; H, 8·1%). *Ethyl butyrylgibberellate,* prepared from ethyl gibberellate with butyric anhydride in pyridine, formed prismatic needles (from ethyl acetate–light petroleum b.p. 40–60°), m.p. 175–176° (Found: C, 67·2; H, 7·5. $C_{25}H_{32}O_7$ requires C, 67·5; H, 7·3%). *Ethyl benzoylgibberellate* was prepared from ethyl gibberellate with benzoyl chloride and pyridine and formed large rhombs (from ether), m.p. 192°

(Found: C, 70·5; H, 6·5. $C_{28}H_{30}O_7$ requires C, 70·3; H, 6·3%). *Phenyl acetylgibberellate.* A mixture of acetylgibberellic acid (39 mg.) and phenol (19 mg.) was treated, at 0°, with trifluoroacetic anhydride (760 mg.) and then stored for 19 h. The resulting solution was evaporated under reduced pressure at room temperature. The residue was treated with water and extracted with ether. The extract was washed with sodium hydrogen carbonate solution, then with water and evaporated. The residual syrup (42 mg.), on treatment with ether–light petroleum (b.p. 40–60°) afforded a solid (16 mg.) which, on crystallisation from moist ether, yielded minute prisms (6 mg.), m.p. 216–219°, of the *phenyl ester dihydrate* (Found: C, 64·6; H, 6·4. $C_{27}H_{28}O_7,2H_2O$ requires C, 64·8; H, 6·4%). *Methyl diacetylgibberellate,*[4] prepared from diacetylgibberellic acid and ethereal diazomethane, formed prisms (from acetone–light petroleum b.p. 60–80°), m.p. 166·5–168·5° (Found: C, 65·1; H, 6·5; OMe, 7·0. $C_{24}H_{28}O_8$ requires C, 64·9; H, 6·4; OMe, 7·0%).[6]

Plant culture and bioassay

Peas (var. Meteor) were germinated in moist sand and then transferred to beakers containing 200 ml. of Long Ashton nutrient solution.[7] Four plants were grown in each beaker, supported on Formica discs, the roots being passed through slits in the discs. After a few days in a greenhouse, when the plant stems had reached a measurable height, the nutrient solution was replaced by fresh nutrient and the experiment commenced. The samples to be tested were applied either in the nutrient solution at 10 μg./ml. or to the leaves in microdrops of ethanol, giving 10 μg./plant. The height of the plants was measured at intervals for 10 days.

Results and discussion

The dose applied was that which in the case of gibberellic acid had been found to induce the maximum increase of growth rate of Meteor pea seedlings during the period of the experiment. Only one dose-level was tested in each case so that the degree of activity was not precisely determined, but it is believed that a reasonably accurate estimation of the activity relative to gibberellic acid was obtained.

Growth rates were fairly constant from 2 to 3 days after application of the compounds until the end of the experiments. In order to compare the results from several experiments they were related to the results for gibberellic acid tested at the same time. Thus, in Table I the results shown were obtained by subtracting the average increment in height of the untreated plants during the 7–8 days commencing 2–3 days after application, from the average increment of the treated plants, in the same period, and expressing these differences as percentages of the corresponding values for gibberellic acid.

Table I

Growth-promoting activity of derivatives of gibberellic acid when applied to dwarf pea seedlings (a) *to the roots in* 10μg./ml. *solution or* (b) *to the leaves at* 10 μg./*plant.*

(The increased growth during 7–8 days of treated plants over that of untreated plants is given as a percentage of the increased growth due to gibberellic acid.)

	Root application	Leaf application
Salts		
NH_4	100	100
Na	95	108
K	100	119
Rb	81	72
Cu	120	81
Ag	136	80
Ca	108	120
Pb	93	103
Mn	114	76
Co	93	91
Cyclohexylamine	100	100
Esters		
Methyl	31	1
Ethyl	43	0
Isopropyl	95	2
Butyl	39	0
Octyl	52	6
Acyl derivatives		
Acetyl	128	89
Butyryl	66	98
Benzoyl	80	103
Diacetyl	75	83
Acyl derivatives of esters		
Methyl acetyl	83	8
Ethyl acetyl	84	2
Isopropyl acetyl	48	4
Butyl acetyl	75	0
Octyl acetyl	2	12
Ethyl butyryl	7	6
Ethyl benzoyl	0	5
Phenyl acetyl	96	not tested
Methyl diacetyl	0	not tested

In general, salts and acyl derivatives showed activities of the same order as that of gibberellic acid itself. Esters and also their acyl derivatives had no significant activity when applied to the leaves in ethanol, but showed moderate to high activity when applied in aqueous solution to the roots. Possible reasons for this discrepancy may be (*a*) that the esters entered the plants more readily by the roots than by the leaves, or (*b*) that they underwent partial hydrolysis during the 10 days the solution stood in the greenhouse. The latter explanation is supported by the results shown in Fig. 1. In this experiment the compounds were applied to the roots of the plants and the stems were measured every 2 or 3 days. The increase in height of the treated plants between each pair of measurements is given as a percentage of the increase of the control plants. The response of the plants to the esters increases with time relative to the response to gibberellic acid. The complete lack of activity shown in Table I by certain acyl esters may be due to low solubility.

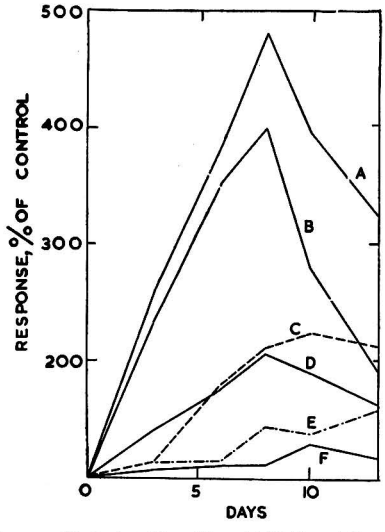

FIG. 1.—*The response of Meteor pea seedlings to gibberellic acid (GA) and its esters expressed as a percentage of the growth of untreated plants*

curve A GA 10 p.p.m.
curve B GA 1·0 p.p.m.
curve C methyl ester 10 p.p.m.
curve D GA 0·1 p.p.m.
curve E ethyl ester 10 p.p.m.
curve F GA 0·01 p.p.m.

The results given above show that the method of application of gibberellic acid derivatives markedly affects their apparent activities in biological assays. Clearly it is necessary to specify the method used when the biological activities of such compounds are being compared.

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Akers Research Laboratories
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EFFECT OF MOISTURE CONTENT ON THE STORAGE OF BRAZIL NUTS

By G. AYERST and D. BUDD

Current practice in the collection, handling and storage of Brazil nuts is described. Laboratory and field methods of determining moisture contents, the relationship between moisture content and relative humidity, and the effect of moisture content on respiration rate have been investigated. In storage experiments, deterioration was slow both at very low and very high moisture contents whereas rather rapid spoilage occurred at intermediate levels. These findings are discussed with reference to current commercial practice and work on other seeds.

Introduction

There has been little published research on the storage of Brazil nuts. Spencer[1] isolated and identified a number of fungi and bacteria from decaying kernels but did not propose methods to prevent decay during storage. Shortly after this Thompson[2] made a series of experiments on their storage and, although the results of this work were apparently not published in detail, his general conclusion was that, for safe storage, the shells of the nuts must be allowed to dry before and during shipment in order to prevent the penetration of fungi.

Bitancourt[3] isolated a number of fungi from decaying Brazil nuts. In a single experiment he found a considerable increase in the number of bad nuts in samples stored at 7° and very high humidities for about 5 months whereas there was no increase in numbers of bad nuts in samples stored at 16–29° and lower but unspecified humidities. He concluded that the main requirement in storage was to maintain the surrounding air at low humidity.

In studying the reasons for the decay of Brazil nuts in storage it was considered essential to know the conditions under which they are stored between harvesting and consumption.

The collection and handling of Brazil nuts has been described in detail by Schreiber.[4] The following brief account of the trade is based on observations made during a visit by one of the present authors (G. A.) to the Amazon area of Brazil in 1956, and in England. Brazil nut trees (*Bertholletia excelsa* Humb. & Bonpl.) grow wild in the forests of equatorial South America. The mature woody fruits, which fall from the trees during the first 5 months of the year, are collected and cut open and the nuts washed in baskets in the rivers, during which process many empty or bad ones are removed by flotation, the sound nuts sinking. The washed nuts are usually stored for several weeks or months by the collector and are then taken by a trader either to an exporting town or to a small trading town. Often several months pass before they are exported in bulk to England, Germany or the United States of America. In the ships the parcels, of between 10 and 100 tons weight and 4–12 ft. depth, are separated by rough timber bulkheads. The holds are ventilated and the top few feet of nuts are turned during the voyage, but some spontaneous heating normally occurs. On arrival in England, usually between May and September, the nuts are cleaned of debris and weighed. They are then kept in well-ventilated floor stores in piles about 4 ft. deep until October to December when they are either sold to the retail trade or re-exported.

Parcels of Brazil nuts always contain a proportion of bad nuts, some of which appear perfectly sound externally and therefore cannot be removed by hand sorting. This proportion is estimated by cracking several samples of 100 nuts from each parcel and is expressed as a percentage, known as the ' crack '. It is usually over 3% on arrival of the nuts in this country, but if it exceeds 10% the parcel is considered to be of inferior quality and fetches a considerably lower price. Because of this it is important to minimise further deterioration.

According to the Board of Trade[5] figures, about 10,000 tons of Brazil nuts valued at about £1,500,000 are imported into this country each year, so that the development of better methods of storage is of considerable commercial importance.

The most obvious change that occurs in the condition of the nuts during storage is a fall in moisture content. Immediately after collection and washing, the nuts contain about 30% of moisture and there is normally little drying during storage in Brazil. Samples taken from twenty different parcels of nuts during loading for export from Manaus in April and May 1956 had a mean moisture content of 28·6% with a range of 20–32%. There is some loss of moisture during shipment and a number of scattered observations have indicated that the moisture content on arrival in England is usually between about 22% and 26%. From this time onwards the rate of drying depends on weather conditions. Samples taken from eight different parcels which had been stored from July to October 1957 had a mean moisture content of 16·3% with a range of 12·8% to 18·8%.

Previous authors[1–3] have emphasised the importance of dryness for the safe storage of Brazil nuts but have not specified safe levels of moisture content. In England and the United States of America nuts containing less than 12% of moisture are normally accepted as being safe for storage (various personal communications).

This paper is mainly concerned with the effect of the moisture content of Brazil nuts on their rate of decay. The relative humidity, moisture content equilibrium, and the effect of moisture content on respiration rate have been studied to help explain the results obtained from the storage experiments.

Experimental

Nuts used in experiments

Brazil nuts are classified by size (small, medium, large and extra large) and by the region in which they are collected or from where they are exported. In all the present experiments large Manaus nuts, the type usually sold for dessert in this country, were used.

A quantity of nuts was obtained in Manaus in 1956 for the first storage experiment, and 560 lb. of freshly imported nuts were obtained in England in 1957 for the second storage experiment. These nuts were also used for relative humidity and respiration measurements.

Determination of moisture content by the drying method

The moisture contents of the shells and the kernels were determined separately. The prepared shells and kernels were dried in tared shallow aluminium vessels for 5 h. at 98° in a vacuum oven (Horwitz[6]) to bring them to virtually constant weight. In early experiments samples of about eight nuts were bulked, the kernels were grated finely and the shells broken into small pieces. In later experiments the moisture content of each nut was determined separately, the kernels being thinly sliced instead of grated. Moisture contents were calculated as percentages of wet weights.

Relationship between kernel, shell and whole nut moisture content

In nuts which have not been recently dried or wetted there is a relationship between kernel, shell and whole nut moisture content. The data summarised in Table I were obtained from a large number of measurements during the storage and other experiments. They are presented in the hope that they will help workers wishing to compare information given only in terms of whole nuts or of kernels. All moisture content figures given elsewhere in this paper are of whole nuts on a wet weight basis.

Table I

Relationship between the moisture contents of the parts of large Brazil nuts

	Moisture content (% of wet weight)								
Whole nut	6	9	12	15	18	21	24	27	30
Shell	9·3	13·9	18·6	22·3	24·7	26·6	28·4	30·1	32·3
Kernel	3·0	4·3	5·9	8·0	10·9	15·2	19·6	24·1	28·2

Moisture content meter

It was frequently necessary to make moisture determinations away from the laboratory and to do this a meter of the electrical resistance type was developed. A pair of spike electrodes mounted on a wooden handle are connected to a 250-V resistance meter of the hand generator type. The spikes are forced through the shell into the kernel of each nut and the electrical resistance between the spikes is measured.

The meter is calibrated at 4·5°, 16·5° and 26·4° and has a range of about 10–24% moisture content. When a sample of twelve nuts is used for determination of moisture content, the 95% confidence limits are about ±0·5% at 10% moisture and about ±1·0% at 22% moisture content. Although this meter is not capable of great accuracy it has been found to be a useful tool for making rapid measurements during the examination of nuts in commercial stores.

Relative humidity/moisture content equilibrium

A knowledge of this relationship is important in understanding the storage behaviour of many commodities and its determination was obviously a necessary part of the present investigation. Measurements were made by allowing samples of nine nuts, which had been slowly dried to suitable levels of moisture content, to reach moisture equilibrium, at constant temperature (20°), with the air in small bottles. The relative humidity of the air in each bottle was then measured either by the cobalt thiocyanate paper method of Solomon[7] or by an electric hygrometer (Negretti and Zambra Ltd.) which was repeatedly recalibrated at known humidities controlled by potassium hydroxide solutions. The moisture content of the nuts was determined by the oven method. In Fig. 1 each point represents one sample of nuts and the line is drawn visually.

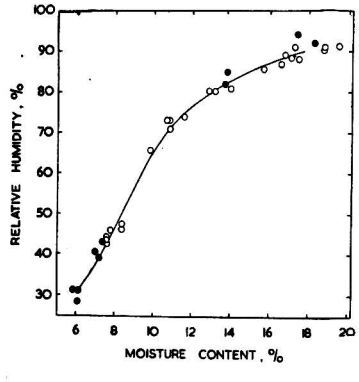

FIG. 1.—*Relative humidity / moisture content equilibrium of whole Brazil nuts*

○ R.H. determined by cobalt thiocyanate paper
● R.H. determined by electric hygrometer

Respiration measurements

The relationship between moisture content and respiration of Brazil nuts is of interest both from the point of view of controlling heating during storage and as an indication of the effect of moisture content on the metabolism of the nuts. Measurements were made on single nuts, as the presence of variable numbers of bad nuts made it difficult to interpret the results of measurements on bulk samples.

Method.—Individual nuts were allowed to respire at a controlled temperature in closed vessels containing potassium hydroxide solution and the fall in the volume of air in the vessels due to oxygen absorption by the nuts was measured. The potassium hydroxide solution acted as the carbon dioxide absorbent, relative humidity controller and manometer fluid. To allow

for errors due to changes in temperature and atmospheric pressure all differences in volume were corrected by the differences occurring in three exactly similar control respirometers which contained no respiring material. Corrections for the changes in pressure in the respirometers due to the fall in level of the manometer fluid were made separately.

The procedure was as follows. Potassium hydroxide solution in equilibrium with the same relative humidity as the nuts was measured into each respirometer so that the remaining air space was the same. An extra 9 ml. of solution was added to each of the three control vessels to allow for the volume occupied by one nut. Nuts conditioned to known moisture contents were placed in the vessels which were then immersed in a water bath maintained at 30 ± 0·5°. The vessels were left to settle for 2 h. before the taps were closed and the potassium hydroxide solution was raised in the side tubes to a suitable level. The levels of the menisci in the side tubes were then noted and further readings were taken at hourly intervals. To avoid fluctuations of temperature the heater of the bath was turned off while readings were being taken. It was found that the rate of fall of volume remained constant throughout each experiment.

Results of respiration measurements

Measurements were made on nuts from the same source as those used in the second storage experiment. Their moisture content was first raised to about 29%; then the nuts were gently dried to a series of moisture contents between 29% and 8% and allowed to equilibrate for at least 2 weeks before respiration measurements were made. Finally the moisture content of each nut was determined by the oven method. Each point in Fig. 2 represents the respiration rate of a single nut. A series of measurements on nuts from a different source gave very similar results.

These results suggest that there are two main levels of respiration rate, a high rate between about 20% and 32% moisture content and a low rate below about 15% moisture content. The transition between these levels is abrupt, but within them changes in moisture content have little effect on respiration.

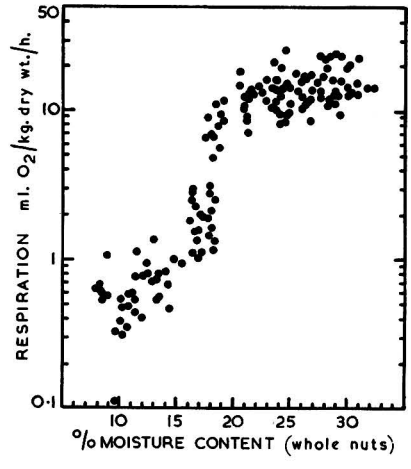

FIG. 2.—*Effect of moisture content on respiration rate of whole Brazil nuts at* 30°

Effect of moisture content on the rate of increase of numbers of bad nuts

Because of the absence of information on the effect of moisture content on the rate of deterioration it was decided to test this point experimentally.

The first experiment was set up in Manaus in May 1956. About 200 lb. of local nuts were divided systematically into 13 groups of just over 600 nuts each and one group was cracked immediately to determine the original percentage of bad nuts. The 12 remaining samples were grouped in pairs. Two pairs were raised to known moisture contents, two pairs were partly dried, one pair was untreated. The sixth pair was ' weathered ', i.e., exposed to the sun and rain for 2 weeks. The samples were all stored in wide-mouthed, 23-litre cans the lids of

which were punctured in several places for ventilation. They were shipped to England where they were stored at a temperature of 20 ± 3°. In December 1956, after 7 months' storage, 300 nuts were randomly selected from each can and cracked. Moisture content determinations were made by the oven method at the same time.

The second storage experiment was set up in England in May 1957. A quantity of nuts, which had been collected directly from a ship, were distributed systematically by small groups into 15 approximately equal samples. Three samples were taken for immediate examination and the rest were distributed by random numbers into four groups which were adjusted to different moisture contents. The samples were stored in enamelled steel bins with loosely fitting lids in an underground chamber in which the summer temperature of about 18° fell to a controlled 15° in the winter and in which the humidity was always above about 70% R.H. One sample at each moisture content was examined after each of the storage periods of two, six and twelve months.

Results of the storage experiments

The degree of deterioration during storage was estimated by the increase in the proportion of decayed nuts.

In the first experiment, there was no deterioration at 29% moisture content and very little at 26% and 23%, but considerable deterioration occurred at 20% and 15% moisture content. The weathered nuts at 20% moisture content deteriorated to the same extent as the corresponding unweathered sample, except that a slightly higher proportion of the nuts became infested with mites. A small degree of drying occurred during storage, but this was insufficient to affect the general results of the experiment.

In the second experiment (Table II), there was slight initial deterioration of the wettest samples but this did not increase after 2 months' storage. In the samples at 23% and 17% moisture content progressive deterioration occurred throughout the storage period, but in the samples at 10–11% moisture content there was no deterioration even after storage for a year. These differences were statistically significant by the heterogeneity of the χ^2 test.

Under the conditions of storage in these experiments there was little deterioration at moisture contents above about 23% or below about 11% moisture content but, in the intermediate range, considerable deterioration occurred. The result of the single test on the effect of weathering suggests that this is not a major factor causing deterioration.

Table II

Effect of moisture content on rate of decay of Brazil nuts (Expt. 2, in England)

Treatment before storage	Storage period, months	Final moisture content, %			Deterioration	
		No. of nuts in sample	Mean	Standard deviation	No. of nuts in sample†	Bad nuts, %
Initial crack	0	24	22·9	2·6	2365	3·85
Soaked in water for two periods of 1 h. on each of 2 days	2	12	27·3	3·1	788	6·2
	6	25	27·9	1·9	789	5·7
	12	26	26·7	2·3	827	6·9
Untreated	2	15	22·8	1·7	804	8·3
	6	23	22·9	1·5	780	11·4
	12	27	23·7	3·0	794	21·7
Dried in unheated and slightly heated air for 9 days*	2	15	17·0	1·2	768	9·4
	6	23	17·2	0·7	781	14·5
	12	24	17·2	1·0	785	21·3
Dried in unheated and slightly heated air for 20 days*	2	18	10·2	0·5	746	4·4
	6	27	10·8	0·8	762	3·4
	12	27	10·8	0·8	788	3·9

† Excluding empty shells

* The temperature of the slightly heated air never exceeded 19°

Discussion

Under the conditions of experimental storage, Brazil nuts kept well at very high or very low moisture contents but deteriorated fairly rapidly at intermediate levels.

Decay is normally due to fungal attack and the investigations of Snow *et al.*[8] and several other workers have shown that only a few drought-resistant fungi can grow at all below about 75% R.H. and that these grow very slowly. Therefore it was not unexpected to find that Brazil nuts at 10–11% moisture content, corresponding to about 70% R.H. are safe for storage. Decay of Brazil nuts would probably be rather slow up to about 13% moisture content (80% R.H.).

It was initially more surprising to find that very moist nuts are also very resistant to decay. However, a variety of living tubers and fruits are normally stored at moisture contents approaching saturation levels. For seeds, dry storage is usually considered to be more convenient and reliable but it is well known that some moist but dormant seeds will survive for periods of several years. For instance, Toole & Brown[9] reported a high percentage germination from some seeds which had been buried in moist soil for 39 years. In laboratory experiments in which lettuce seeds were kept at 85–90% R.H. and 30°, Toole & Toole[10] found that soaked seeds maintained their viability far better than unsoaked seeds. Barton[11] showed that seeds of *Amaranthus retroflexus, Impatiens balsamina* and *Rumex obtusifolius* could be maintained in a dormant but viable condition for over a year when saturated with water.

For successful storage at high moisture contents a seed must remain dormant and resist attack by fungi and other organisms. Brazil nuts have a prolonged dormancy which is not readily broken by changes in temperature or moisture content, or by mechanical shock. Their robust shells are an effective barrier to insects and mites and prevent bruising or surface damage to the kernels which would encourage fungal attack. Our experience indicates however that their main resistance to fungal penetration occurs at the surface of the kernel. The outer layer of kernel cells is covered with a thick cuticle which is presumably an effective barrier to fungal penetration. Even if this outer layer of cells is broken, in nuts with high moisture content, healing often occurs and this is associated with suberisation of cell walls and the presence of dark brown inclusions in other cells. It is not unusual to find nuts in which the shell and a small part of the kernel have been cut away, presumably during the opening of the fruit, and in which healing has been so effective that there is only a small scar on the kernel.

Penetration by fungi appears therefore to be prevented by the ability of the healthy living cells of the kernel to resist attack. If these cells are wilted, as in partly dried nuts, it is reasonable to suppose that they offer less resistance to penetration, and they would certainly be less capable of healing wounds. The range of moisture content at which resistance to fungal attack is effective is about that at which the respiration rate of the nuts is at its highest level while the rapid fall in respiration rate with falling moisture content between about 20% and 15% is correlated with the maximum susceptibility to decay which occurs in this region. This supports the idea that there is a strong connexion between the metabolism of the nut and its resistance to decay.

If it is accepted that the resistance to decay of moist nuts is dependent upon the healthy condition of the living tissues, it follows that anything capable of damaging these tissues could encourage decay. Exposure to excessively high temperatures or depletion of oxygen supply could cause such damage and these conditions may both occur in some large bulks of actively respiring nuts.

At present Brazil nuts are usually kept very moist during storage in Brazil but during the first few months of storage in England the moisture contents fall to levels at which the nuts are very susceptible to decay. This probably accounts, at least partly, for the deterioration which occurs in parcels of nuts stored in this country. It seems that this deterioration could be reduced either by maintaining the nuts at above 25% moisture content, or by drying them from this level to about 12% moisture content over a period of not more than a few weeks.

Storage at high moisture content is attractive in that the nuts appear to be fresher and more creamy, but it can probably be used for only part of the total imports. Some development work would obviously be necessary before this method could be used commercially in this country.

Dried nuts can apparently be stored indefinitely without special attention, but there is a considerable weight loss during drying and this is very important with such a high-priced

commodity. Drying is at present carried out on a relatively small scale in Brazil but rather high temperatures are used and it is said that there is some deterioration in flavour. There seems little doubt that a satisfactory drying method for use in this country could be developed and, if so, drying would seem to be the better method for treating the nuts intended for re-export or pre-packaging.

Acknowledgments

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PRODUCTION OF HEAT DURING FERMENTATION OF CACAO BEANS

By R. H. KENTEN* and B. D. POWELL†

The heat produced during the fermentation of cacao beans could arise from the action of the bean enzymes as well as from the activities of micro-organisms on the pulp which surrounds the beans. Experiments to ascertain the relative importance of each of these sources have shown that most of the heat produced arises from the activity of micro-organisms in the pulp.

Introduction

Fermentation[1] is the first step in the processing of cacao. After removal from the pod the beans are put into boxes, baskets or piled into cone-shaped heaps. During this stage the pulp surrounding the bean becomes contaminated with micro-organisms. The temperature of the mass of beans rises and at the same time the micro-organisms multiply rapidly. If the

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temperature rise is too slow or a sufficiently high temperature is not reached, a product containing germinated and unfermented beans is obtained. Since this is undesirable as cocoa and chocolate of good quality cannot be made from it, it is important to know on what factors the production of heat depends. Heat can arise from the activities of micro-organisms on the pulp and from enzyme actions in the pulp, testa and cotyledons of the beans themselves. There has been some speculation[2] on the possible importance of these separate sources of heat in the fermentation process but no work has been described which differentiates between them experimentally. The results of the present work strongly suggest that most of the heat is produced by the action of micro-organisms on the pulp.

Experimental

Methods

Beans from ripe Forastero Amelonado pods were used throughout, the pods being opened 3–5 days after harvesting. No attempt was made to avoid chance contamination except in the special experiments, but all the work was done under clean conditions and the pods were thoroughly washed with tap water before opening. The beans or bean parts were packed into vacuum flasks, a thermometer positioned in the middle of the beans* and the mouth closed with a plug of non-absorbent cotton wool. The assembly was then placed in a constant-temperature room or incubator (usually at 30°) and the rate of increase in temperature of the bean mass was followed. Either 1-litre flasks holding 200–250 beans or 250-ml. flasks with 50–60 beans were used.

Washed beans

Beans were largely freed from pulp by shaking with a few soft rubber bungs and water for 2–3 h. with frequent changes of water. The beans were finally rolled on filter paper to remove excess moisture before being packed into the flasks.

Skinned beans

The pulp and testa were removed from the beans with a scalpel and the skinned beans washed with several changes of distilled water. After excess water had drained off the surfaces of the beans were dried with filter paper.

Pulp plus testa

Beans were skinned as described above and the pulp plus testa well mixed with a weight of small rubber bungs approximately the same as that of the cotyledons removed. This was done to replace the cotyledons with a bulk of inert material having a similar specific heat. Unfermented cacao cotyledons are approximately one-third fat and one-third water, the rest being largely carbohydrate and protein. Although their specific heat has not been reported, it is unlikely to be widely different from that of rubber.

Sterile beans

Whole beans were surface-sterilised by immersion for 5 min. in 1% w/v mercuric chloride and allowed to drain in a filter funnel.

Whole beans with diminished contamination by micro-organisms

The work was done in a small room free from draughts, and the instruments, cotton wool and thermometers were sterilised before use by heating for 20 min. at 15 lb. pressure in a steam autoclave.

Pods free from external blemishes were swabbed with 90% ethanol, opened with a knife and the beans stripped from the placenta with a large pair of forceps directly into a vacuum flask, which had been previously sterilised by rinsing firstly with 90% ethanol and then three times with sterile water. Finally the mouth of the flask was closed with a plug of cotton wool and a thermometer positioned in the middle of the beans.

* The temperature was not uniform throughout the flasks but the differences were less than 2°

Effect of aeration

One-litre vacuum flasks were packed with whole beans or skinned beans and a glass tube was positioned so that it reached the bottom of the flask. By this means a stream of washed and filtered air (200 ml./min.) was passed through the beans for 5 min. four or five times every 24 h.

Results

Effect of aeration

Although there is evidence[3] that oxygen assists the rate of heating of a normal cacao fermentation, under the conditions used in the present work, increasing the aeration did not bring about any appreciable increase in the rate of heat production, either by whole beans or skinned beans. Presumably sufficient oxygen was present initially or it diffused in during the experiments. All subsequent experiments were therefore made without forced aeration.

Washed beans and skinned beans

The effect on the rate of production of heat that was brought about either by washing off most of the pulp, or by removing the whole of the pulp together with the testa, is shown in Fig. 1. Both treatments reduce the rate of heating considerably; after 50-h. incubation the treated beans were only 2°–5° above the ambient temperature, whereas the untreated beans were 13°–15° above it. At the end of the experiment more than 90% of the washed beans had germinated and they were still viable. The control beans were all dead and none had germinated, presumably because the rapid heating killed them before germination could take place.

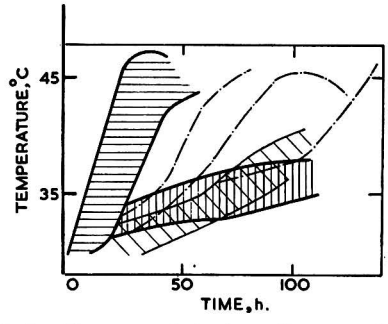

FIG. 1.—*Thermogenesis in fermenting cacao beans*

Ambient temperature, 30°

The individual temperature-time curves have been plotted where whole beans with diminished contamination by micro-organisms were used as these curves differed widely and only three experiments were made. The curves for the other more numerous experiments did not show such large variation. Consequently, for the sake of clarity, these results are shown as areas which contain all the individual results. Thus, all the experimental points obtained with control whole beans lie in the area hatched with horizontal lines.

whole beans | skinned beans | washed beans

·—·— special treatment whole beans

The rate of respiration[4] and hence the rate of heat production increases with the onset of germination and continues at a high level throughout germination. The greatest production of heat from enzymic processes in the cacao bean itself would therefore be expected under the conditions which lead most readily to germination. These conditions must have been approached in the experiments with the washed beans where germination was virtually complete and yet the rate of heating was slow.

The results suggest therefore that most of the heat produced when cacao is fermented arises from processes in or on the pulp, a suggestion supported by the results from the experiments with the testa and pulp of beans mixed with rubber bungs, which heated up at the same rate as whole beans.

Respirometer experiments

The heat produced by the pulp could arise from the activities of pulp enzymes as well as those of micro-organisms. Experiments in the Warburg respirometer with pulp removed from the beans under aseptic conditions showed that the respiration of pulp itself was much less than that of washed or skinned beans, suggesting that the heat produced by the activity of the pulp enzymes would be small. Results of respiration experiments are shown in Table I.

Table I

Results of respirometer experiments

	(μl. of gas/g. fresh weight/h.) Oxygen	Carbon dioxide	No. of experiments	Error
Skinned beans	65	68	8	±20%
Washed beans	44	42	4	±20%
Pulp	13	8	6	±30%

Beans with diminished microbial contamination

If most of the heat produced by whole beans arises from the activities of micro-organisms on the pulp, and if the amount of initial contamination could be lowered, a longer lag period (for the multiplication of the micro-organisms) would be expected before the phase of rapid heating began. Three experiments were made in which beans were removed from the pod under the conditions likely to diminish the contamination by micro-organisms and, in each, the beans likely to be less contaminated showed a longer lag period than the control (Fig. 1).

Surface-sterilised beans

Whole beans, the surface of which had been sterilised by treatment with mercuric chloride, heated up at a slower rate than skinned beans. The beans remained viable after the treatment but in view of the known toxic effects of mercury ions on enzyme systems such experiments do not differentiate between the contributions to the total heat production from the pulp enzymes and that from the activities of micro-organisms on the pulp. However, because of the low respiration rate of aseptic pulp they do suggest that the mercuric chloride treatment prevents the production of heat because it inhibits the growth of the micro-organisms. These experiments were done with the small vacuum flasks at an ambient temperature of 35°.

Dead beans

Working with small vacuum flasks at an ambient temperature of 35°, whole beans were used which had been treated for 2 h. in a steam oven and allowed to cool before being packed into the flasks. These heated up at the same rate as the untreated beans. Since the heated beans were dead and most of the enzyme systems had been inactivated, the production of heat by these systems must have been negligible and it is reasonable to assume that all the heat was produced from the activity of micro-organisms on the pulp.

Discussion

All the evidence obtained in the present work points strongly to the conclusion that most of the heat produced during fermentation arises from the activity of micro-organisms on the pulp. It is of interest that the production of heat, which causes serious damage to such products as grain and soya-beans,[5] when they are stored under too moist conditions, is also due largely to the activity of contaminating micro-organisms, and that the enzyme activity of the seeds themselves plays only a minor rôle.

The present work suggests that the success of a fermentation by the usual methods is initially dependent on chance contamination by micro-organisms, and variation in the extent of this contamination may in part account for the large differences in the rate of heating sometimes observed between fermentations made under apparently identical conditions. In West Africa the authors have never known a properly set up fermentation to fail to heat, but Bridgland[6] has encountered ' dead fermentations ' in New Guinea. Insufficient or unsuitable contamination by micro-organisms could account for this rare phenomenon.

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RESIDUES OF *OO*-DIMETHYL *S*-(*N*-METHYLCARBAMOYLMETHYL) PHOSPHOROTHIOLOTHIONATE (DIMETHOATE) IN SPRAYED CROPS

By E. D. CHILWELL and P. T. BEECHAM

A method for the determination of residues of the insecticide *OO*-dimethyl *S*-(*N*-methylcarbamoylmethyl) phosphorothiolothionate (dimethoate) in crops is described. Radiochemical and biochemical investigations indicate that the method can be applied to growing crops without interference from metabolites in the plant derived from dimethoate. The limit of sensitivity of the method is about 5 μg. of dimethoate, equivalent to 0·1 p.p.m. in the plant tissue by the method described. Harvest residues found in many British and overseas crops 1–3 weeks after spraying with dimethoate are given.

Introduction

OO-Dimethyl *S*-(*N*-methylcarbamoylmethyl) phosphorothiolothionate is a systemic organophosphorus insecticide of comparatively low mammalian toxicity.[1, 2] It is the active ingredient of the commercially available insecticide ' Rogor* 40 ' which is a water-miscible formulation containing the equivalent of 3·0 lb. of active ingredient (a.i.)/Imperial gallon. The accepted common name in North America is ' dimethoate ', but no common name has yet been adopted in this country. For convenience in this work the name ' dimethoate ' is used to denote the pure chemical, $(CH_3O)_2PS{\cdot}S{\cdot}CH_2{\cdot}CO{\cdot}NH{\cdot}CH_3$, and ' Rogor ' to denote the formulated material.

Dimethoate is a white colourless crystalline solid, with m.p. 49–50°, vapour pressure approx. 1×10^{-4} mm. Hg at 25° and solubility in water 2·5 g./100 ml. at 25°. Partition coefficients for various solvents at 25° (most favoured phase first) are : water/light petroleum (b.p. 80–100°) 23·5 ; chloroform/water 32·5 ; water/carbon tetrachloride 2·2. It is hydrolysed by aqueous alkali ($K = 6{\cdot}8$ min.$^{-1}$ moles^{-1} at 25°) with half-life approximately 1 min. in 0·1N-NaOH and is relatively stable in neutral and weakly acid solutions. The compound slowly decomposes on storage ; initially neutral it gradually becomes acidic.

* ' Rogor ' is the registered trade mark of the Montecatini Co.

As a proprietary formulation, Rogor was introduced in 1956 for the control of fruit flies on olives and cherries in the Mediterranean area. Extensive work carried out in the United Kingdom and in other countries has shown that Rogor is effective against a wide variety of pests, especially aphids and mites on field, fruit and horticultural crops.[2] Its use on a food crop necessitates the determination of residues persisting in the edible crop in order to demonstrate the absence of any detectable toxic hazard to the consumer.

Determination of residue

Methods for the determination of dimethoate residues in olive oil by solvent extraction and phosphorus determination,[3] and in cherries by colorimetric determination of methylamine liberated on acid hydrolysis[4] have already been published. A bioassay method using *Drosophila melanogaster* has also been successfully employed for residue determinations.[5] For the investigation of possible residues in a wide variety of crops a method known to be applicable to many different types of sample was desirable. The method of Heath *et al.*[6] has been used extensively in this laboratory, depending on the microdistillation of the residue remaining after evaporation of a chloroform extract. It was known to be applicable to the determination of residues of *OO*-diethyl *S*-(*N*-isopropylcarbamoylmethyl) phosphorothiolothionate, which is closely related to dimethoate, and the possibility of applying the method to dimethoate residues was investigated. By modification of the extraction procedure, and an increase in the temperature of distillation, the technique was found to be suitable. The analytical method now in use is as follows.

Apparatus.—The dimensions of the microdistillation apparatus used are the same as described by Heath *et al.*,[6] but the lower surface of the cold-finger condenser is ground to give a rough surface for greater area.

Procedure.—Take 250 g. of sample and macerate with approximately 250 ml. of water and acidify to pH 4·0 with acetic acid (10%). Filter through a Buchner funnel. Repeat the maceration at pH 4·0 twice, the extract being filtered off each time. Centrifuge the combined aqueous extracts and make up to 1 litre. Take 200 ml. of clear extract, neutralise to pH 7·0 with 2N-NaOH and extract twice with an equal volume of chloroform. Emulsions are rarely encountered if the volume of chloroform is equal to, or greater than, the volume of the aqueous extract. Combine the chloroform extracts and filter (Whatman No. 1 paper). Distil off the chloroform at 60° using a water-pump until approximately 10 ml. of extract remains. Transfer to the special microdistillation flask and evaporate to dryness at as low a temperature as possible, with gentle suction to remove the final traces of solvent. Insert the cold-finger condenser and evacuate to a pressure of 1 mm. Hg. Maintain the temperature of the lower third of the flask at 200° for 30 min. by placing in an oil bath or heating block. Take out of the block, release the vacuum, remove the cold finger carefully and rinse the lower part into a small beaker with chloroform. Transfer the washings to a 50-ml. Kjeldahl digestion flask, add 5 ml. of distilled water and 2 ml. of 72% perchloric acid, add a small glass bead and heat gently to remove chloroform. When the solvent has evaporated, add 5 drops of HNO_3 and heat to white fumes. Cool, add a few ml. of distilled water and heat again to white fumes. Cool, dilute with 20 ml. of water and transfer to a 25-ml. separating funnel. Determine the phosphorus content colorimetrically by the modified Berenblum & Chain method described by Heath, or any suitable colorimetric method.

The phosphorus content is calculated from a calibration curve obtained with potassium dihydrogen phosphate, and

$$\text{Dimethoate} = P \times 7{\cdot}40$$

With a Unicam SP 600 spectrophotometer and a 1-cm. cell, an optical density of 0·50 was given by 61 μg. dimethoate. The limit of sensitivity of the method is about 5 μg. of dimethoate, equivalent to a residue of 0·1 p.p.m. in the plant tissue.

Extraction of dry samples

The method has been found applicable to all the fresh vegetable and fruit crops analysed. In a few crops of high solids content such as dried hops, tobacco and tea, the extraction described

is not sufficiently selective and the method breaks down at the distillation stage due to the excessive amount of chloroform extractives present.

The methanol extraction of Field & Laws[7] was adopted for dried hops, the aqueous methanol being washed with a small volume of carbon tetrachloride in addition to light petroleum used by Laws.

Tobacco and tea were mixed with anhydrous sodium sulphate and extracted with a mixture of acetone and light petroleum (b.p. 40°–60°) (1 : 4). On addition of an equal volume of water the dimethoate partitions into the aqueous acetone leaving most of the interfering extractives in the light petroleum.

Efficiency of extraction procedure

Small lettuce seedlings were sprayed with ^{32}P-labelled dimethoate as a 0·2% solution in water containing Lissapol NXA as a wetting agent. The plants were harvested 1 and 4 days after treatment and extracted by maceration with water as described above. The extracted plant debris was digested with HNO_3 and H_2SO_4 to destroy organic matter. The radioactivity of the water extract and of the digested plant debris was determined with a liquid-sample Geiger–Müller tube and scaling unit. The proportion of radioactivity extracted by three water macerations was 95·8% and 96·3% of the total found in water and digested plant debris. Therefore at least 96% of the dimethoate residue in the plant would be extracted by water.

Specificity

The method registers all phosphorus compounds extractable by chloroform from water which are sufficiently volatile to distil under the conditions described. Experience has shown that these include very few natural products, but will include residues of schradan, demeton, demeton-methyl and other organophosphorus insecticides if these are present.

Blank on untreated crops and recovery experiments

The 'blank' obtained by carrying out the whole analytical procedure on a sample not treated with insecticide has been obtained on 24 crops. The average value on all crops is 0·13 p.p.m., the range of individual results being 0·00–0·27 p.p.m.

The recovery of a known amount of dimethoate added to a plant extract is shown in Table I. The average recovery is 75% but the Table indicates that recovery may vary significantly from crop to crop.

Table I

Recovery of dimethoate added to different crops

Crop	Level of added dimethoate, p.p.m.	Recovery (mean), %	Crop	Level of added dimethoate, p.p.m.	Recovery (mean), %
Sugar beet	1·0	82·9	Plums	1·0	63·0
Brussels sprouts	1·0	75·2	Pears	1·0	62·4
Lettuce	1·0	85·2	Peaches	1·0	75·1
Lettuce	0·5	99·8	Potatoes	1·0	84·0
Cabbage	1·0	80·1	Potatoes	0·2	70·0
Cabbage	0·5	69·0	Field beans	1·0	77·6
Apples	1·0	71·1			

Relationship of residue determined to toxic hazards to the consumer

The result of dietary feeding trials on groups of 20 male rats for 12 months and other direct data on human ingestion, has led to the view that a residue of 2 p.p.m. of dimethoate in a harvested crop for human consumption would be harmless.[1]

The analytical values obtained for residues in a crop treated with insecticide must be related to toxicological studies on the chemical before an opinion can be given on the safety of the crop for human consumption. One important consideration is that the analytical residue should represent the total hazard to the consumer. This is perhaps particularly important with systemic insecticides which are absorbed into and broken down by the plant, and the possibility arises of metabolites being produced which are not detected in the analysis. The possibility

of any stable non-ionic metabolites being formed during the life of dimethoate in the plant was investigated by partition chromatography with ^{32}P-labelled insecticide, as described by Heath *et al.*[8]

Lettuce and broad bean seedlings harvested 5 days after spraying with ^{32}P-labelled insecticide were thoroughly extracted with chloroform and the concentrated extract chromatographed on a kieselguhr column with a mixture of chloroform, trichloroethylene and carbon tetrachloride (3 : 7 : 10) as the moving phase. The major peak, containing 97% of the radioactivity, was found in the first 100 ml. of eluent with a minor peak containing the remaining radioactivity between 300 and 500 ml. of eluent. The major peak partitioned strongly in favour of chloroform from water, whilst the second minor peak partitioned equally between chloroform and water. Both peaks showed an increase in anti-cholinesterase activity compared with that of the solution originally applied. Both have subsequently been demonstrated to be present in an extract from mammalian liver following administration of dimethoate. Santi & Pietri-Tonelli[9] have prepared the oxygen analogue of dimethoate and shown by solubility and infra-red data that it is identical with a metabolite extracted from plants.

Edson[10] has suggested that the most expeditious method of proving that metabolites do not invalidate the analytical residue results is by animal feeding experiments. An experiment was therefore carried out to compare the effect of feeding animals with dimethoate as a crop residue determined by analysis and as a pure chemical fed at the same rate. Groups of guinea-pigs were fed daily for 3 weeks on large known amounts of fresh lettuce, the dimethoate residues in which were maintained by repeated excessive applications at 130–190 p.p.m. as shown by chemical analysis. The animals thus ingested a known daily dose of dimethoate as a residue in a treated crop. Other guinea-pigs were then given the same daily dose of dimethoate orally as a propylene glycol solution of the pure laboratory material, for 3 weeks. The blood cholinesterase activity assessed after 1, 2 and 3 weeks, respectively, on heart-blood samples were (as % of normal activity) 49, 30 and 20% for pure dimethoate, and 48, 19 and 23% for dimethoate ingested as crop residue. Thus the anticholinesterase effect of dimethoate was quantitatively similar for the two sources of dimethoate. It was concluded therefore that the analytical method gave a valid estimate of the total anticholinesterase hazard to the consumer.

Harvest residues

The residue present in the edible portion of a sprayed crop at harvest is the critical factor in assessing the hazard to the consumer. Such residues have been determined on a wide range of crops grown in this country, and on several of the more important crops from overseas.

The data given in Table II have been obtained over the past two seasons from experimental trials with Rogor on home-produced crops. Residues found in some important overseas crops are given in Table III. In all cases residues were determined after spraying, but since Rogor is also effective when applied to the soil some residues were determined using this method of application. The samples taken represented the mature edible crop in a suitable condition for marketing. Cabbages and lettuce were hearted and about four outer leaves were retained in the sample. Root crops were washed to remove soil, but otherwise the samples were analysed without being cleaned. The residues reported have been corrected for the ' blank ' on the control samples of untreated crop and for incomplete recovery (75%).

In most of the garden crops for which results are given in Table II, the residues after soil application, and also after application as a drench from a watering can, were determined. The residues were found to be similar to those on sprayed crops, except in a few cases where residues of 3–4 p.p.m. were found after drenching, probably due to the large volumes of liquid required to give adequate cover of the crop by this method.

In some fruit crops a remarkable difference was observed in the residues found in the fruit and in the leaves. Residues in the fruit are 0·5–1·0 p.p.m., at 1–3 days after spraying, whilst leaves contain 10–20 p.p.m. In one experiment on apples the values found in leaves and fruit, respectively, were 14·1 and 0·5 p.p.m. at 4 days, and 6·9 and 0·2 p.p.m. at 10 days after application. A similar observation has been reported for demeton.[11] In green vegetable crops more residue was found in the outer leaves than in the edible parts of the crop. Brussels sprouts

analysed 8 days after spraying contained 4·9 p.p.m. of dimethoate in the leaves, and 0·7 p.p.m. in the sprouts. Cabbages examined 7 days after late-autumn spraying contained 2·4 p.p.m. in the outer leaves, 1·4 p.p.m. in the wrapper leaves and 0·4 p.p.m. in the edible heart. Such differences are probably due to the greater surface area/mass ratio for open foliage than for fruit or compact foliage.

In citrus fruits the peel and pulp were analysed separately, and most of the residue was found in the peel. In oranges harvested 14 days after spraying residues found were 2·33 p.p.m. in peel and 0·3 p.p.m. in pulp at 12·8 oz./100 gal., and 2·9 and 0·7 p.p.m. at 25·6 oz./100 gal. In apples comparable figures obtained were—peel 1·9 p.p.m., peeled apple 1·1 p.p.m. and for peaches 3·1 and 1·8 p.p.m. A higher concentration of insecticide in the skin seems to occur in fruit generally, but this is most marked in the citrus species.

Table II

Harvest residues in U.K. crops

Crop	Application		Date of application	Interval from last application, days	Dimethoate residue, p.p.m.
	Concn. of spray, oz. a.i./100 gal.	Rate, gal./acre			
Sugar beet	12·8	High volume	18.6.57	100	<0·1
	38·4	Low volume	18.6.57	100	0·2
	6·4	High volume	5.7.57	77	<0·1
	3·2	High volume	5.7.57	77	<0·1
	51·2	Low volume	30.7.57	48	<0·1
	38·4	Low volume	18.6.57	100	0·2
	6·4	High volume	5.6.58	131	0·2
Brussels sprouts	25·6	100	16.8.57	12	1·1
	25·6	100	16.8.57	19	0·3
	24·6	100	10.10.57	8	0·7
	25·6	100	10.10.57	15	0·1
	25·6	100	22.11.57	7	0·5
	6·4	100	21.10.58	7	0·2
	6·4	100	21.10.58	14	0·2
Cabbage (summer)	6·4	100	26.8.58	7	0·1
	6·4	100	26.8.58	14	0·7
	6·4	100	26.8.58	21	0·2
Cabbage (autumn)	6·4	100	11.11.58	7	0·3
Cauliflower	6·4	100	27.8.58	14	0·5
	6·4	100	11.9.58	7	1·3
Kale	8	100	26.8.58	13	0·3
Peas (shelled)	6·4	100	17.7.58	20	0·1
	6·4	100	23.7.58	7	0·3
	6·4	100	23.7.58	21	0·5
Lettuce	6·4	50	12.6.57	13	0·1
	6·4		15.7.58	7	0·3
	2·0	200	5.5.59	7	1·1
Lettuce (greenhouse)	2·0	200	5.5.59	7	0·9
Broad beans	9·6	100	10.7.58	7	0·5
Runner beans	16	100[a]	24.7.58 20.8.58	7	0·7
Dwarf beans	16	100[a]	20.8.58	7	0·4
Beetroot	16	100[a]	20.8.58	7	0·9
Carrots	16	100[b]	30.6.58 24.7.58 20.8.58	14	0·6
Onions	16	100[b]	30.6.58 24.7.58 20.8.58	14	0·2

Table II (*cont.*)

Crop	Application: Concn. of spray, oz. a.i./100 gal.	Application: Rate, gal./acre	Date of application	Interval from last application, days	Dimethoate residue, p.p.m
Potatoes	16	100[b]	30.6.58	14	0·1
			24.7.58		
			20.8.58		
	Seed treatment		20.4.58	150	<0·1
Turnips	16	100[a]	30.6.58	14	0·4
			24.7.58		
Strawberries	12·6	100	13.6.57	20	0·2
	6·4	100	8.7.58	7	0·3
Blackcurrants	8	100	14.7.58	7	0·1
	8	100	14.7.58	14	0·4
Apples	6·4	50	3.7.58	7	0·4
	6·4	300	2.7.58	8	1·1
	6·4	250	3.7.58	6	0·3
	6·4	250	15.7.58	7	0·6
Cucumber (greenhouse)	8	100 (sprayed to run-off)	15.7.58	6	0·3
	Soil application 4·3 oz. a.i./acre		3.7.58	4	0·6
			3.7.58	12	0·8
			3.7.58	18	0·3
			3.7.58	24	0·1
Tomatoes (greenhouse)	6·4	100 (sprayed to run-off)	26.8.58	7	0·4
	Soil application 4·3 oz. a.i./acre				
	first application		3.7.58	4	0·1
				7	0·1
	second application		31.7.58	5	0·4
				7	0·6

[a] 2 applications [b] 3 applications

Table III

Harvest residues in overseas crops

Country of origin	Crop	Application rate	Interval from last spray to harvest, days	Dimethoate residue, p.p.m.
South Africa				
January, 1958	Apples	0·036% a.i., to run-off	9	0·3
			14	0·2
			21	0·3
			31	0·1
January, 1958	Plums	0·018% a.i., to run-off	3	0·2
January, 1958	Pears	0·018% a.i., to run-off	3	0·2
			8	0·4
			13	0·1
			20	0·3
January, 1958	Peaches	0·053% a.i., to run-off	8	1·1
			20	0·4
			30	0·1
January, 1959	Potatoes	6 oz. a.i., 130 gal./acre§	14	0·1
February, 1959	Kaffir corn	8 oz. morgen	17	0·1
			21	0·2
		12 oz. morgen	17	0·1
			21	0·1
		15 oz. morgen	17	0·1
			21	0·2

§ 7 sprays at weekly intervals

Table III (*cont.*)

Country of origin	Crop	Application rate	Interval from last spray to harvest, days	Dimethoate residue, p.p.m.
Cyprus				
July, 1958	Oranges (peeled) (Shamouti)	0·2% a.i., 25–30 l./tree	14	0·7
			21	0·7
		0·1% a.i., ,, ,,	14	0·3
			21	0·5
January, 1959	Oranges (peeled) (Valencia)	0·133% a.i., 30 l./tree	14	0·6
			21	0·7
October, 1958	Lemons (peeled)	0·133% a.i., 40 l./tree	14	0·4
			21	0·1
October, 1958	Grapefruit (peeled)	0·133% a.i., 3 l./tree	14	0·1
			21	0·1
Kenya				
April, 1958	Oranges (peeled)	0·2% a.i., 3 l./tree	18	0·1
May, 1958	Coffee beans	0·22% a.i., 40 gal./acre	2	0·5
Canada				
September, 1958	Potatoes	Spray 12 oz. a.i., 100 gal./acre	25	0·1
			55	0·1
		Dip 1% a.i. at planting	126	0·1
September, 1958	Wheat	Spray 1 lb., 30 gal./acre	58	0·2
		Seed treatment 5% dust	114	0·1
Canary Islands				
January, 1959	Oranges (peeled)	0·014% a.i., 1·7 l./tree	16	0·3
Australia				
January, 1959	Apples	0·04% a.i., 12 pints/tree*	7	1·2
	Nectarines	0·04% a.i., 12 pints/tree*	7	1·9
	Peaches	0·04% a.i., 12 pints/tree*	7	2·0
January, 1959	Oranges (peeled)	0·1% a.i., 2 gal./tree†	14	0·6
New Zealand				
April, 1959	Apples	12·8 oz. a.i./100 gal. to run-off	10	0·9
Switzerland				
September, 1957	Apples	0·018% a.i., to run-off	4	0·5
			10	0·2
	Grapes	0·018% a.i., to run-off	4	1·6
			10	0·7
			20	0·3

* 3 sprays at 7-day intervals † 2 sprays at 1-month intervals

Discussion

The application rates used in these experiments were in many cases greater than the recommended dose for efficient control of aphis to allow for accidental overdosage in spraying, but the residues found by analysis are below the 2 p.p.m. level suggested as a safe limit in edible commodities.

In sugar beet the spray was applied at both high and low volume, and on beans and root crops the residues were determined after two or three applications. Potatoes received from South Africa had been sprayed weekly for 7 weeks with no detectable increase in residue. On brassicas, spraying was continued until late autumn without any unacceptable residue being found in the edible hearts or sprouts.

During the 1958 season residues were determined on 23 home-produced crops treated by spray, soil application and drenching the plant from a watering can. In Table IV the results have been grouped according to the residue found and the interval between application and harvest, irrespective of the crop or the method and date of application.

Table IV

Summary of results for dimethoate residues on home-produced crops

Interval between last application and harvest	No. of samples	Percentage of samples with residues between limits (p.p.m.)				
		<0·5	0·5–1·0	1·0–1·5	1·5–2·0	>2·0
7 days (range 1–7)	65	44·6	33·8	12·3	6·2	3·1
14 days (range 9–17)	77	70·1	23·4	5·2	1·3	—
21 days (range 18–28)	44	84·1	11·4	4·5	—	—
Total samples	186	64·5	24·2	7·5	2·7	1·1

At 7 days after application, dimethoate residues are below 2 p.p.m. in 97% and below 1 p.p.m. in 78% of the samples, a level which is insufficient to cause cholinesterase depression in mammals. Entomological investigations have shown that the plant remains toxic to aphids for 10 to 21 days after spraying, and for longer periods with some fruit-boring flies. This observation is in agreement with that of Dauterman *et al.*[12] who showed that the LD_{50} of dimethoate to houseflies is more than 1000 times greater than to rats, and with unpublished studies implying that locusts lack the ability to break down one of the metabolites of dimethoate and are, in consequence, more susceptible than mammals. Santi & Pietri-Tonelli[9] state that the oxygenated metabolite of dimethoate present in traces in plants is more acaricidal than is dimethoate. The animal feeding experiment described in this paper indicates that residual dimethoate and its metabolites in lettuce leaves are no more toxic to mammals than is pure dimethoate.

During the course of this work, analyses were made on the same crop grown and treated in several countries. For example, apples have been analysed after spraying in the United Kingdom, Switzerland, South Africa and Australia. The residues at comparable intervals after spraying were all in the range 0·1–1·2 p.p.m. This is interesting in considering the effect of climate on residues, since, at least with apples, a treatment which effectively controlled insect infestation carried out in four entirely different locations, but applied at the same stage of development of the fruit, i.e. nearing maturity, gave very similar residue values. Similar agreement in results was seen in experiments in two countries on potatoes, oranges and peaches. The deduction is that climate itself is not a major factor in the rate of fall-off of dimethoate residues in growing crops.

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KAFFIRCORN MALTING AND BREWING STUDIES. IV.*—The Extraction and Nature of the Insoluble Amylases of Kaffircorn Malts

By L. NOVELLIE

The α- and β-amylases produced by the germination of kaffircorn may be either soluble or insoluble depending on the variety of the kaffircorn. The insoluble amylases occur chiefly in malts from ' birdproof ' kaffircorn and sweet sorghum. The amylases of these varieties, unlike the bound amylase of barley, are active in the insoluble state. Proteins, peptone, ethylenediamine, histidine and compounds containing the group —N·C(X)·N— (where X is not oxygen) effect maximum liberation of the insoluble amylases. Partial solution is obtained with salts, surface-active agents and certain basic substances. The release of the amylases is not a proteolytic process but possibly a desorption from the surface of an insoluble protein.

Introduction

The application of the aqueous extraction method for determining diastatic power[1] to malts made from ' birdproof ' kaffircorn gave very low values, usually less than 5 K.D.U.†/g. Nevertheless these malts were as active in thinning and saccharifying mashes as those of diastatic power 25–30 K.D.U./g. made from other varieties of kaffircorn. From this it was concluded that the amylases of birdproof kaffircorn malts were insoluble in water but were active during the mashing operation. This insolubility bore, at first sight, some resemblance to that found with barley grain amylase. Further investigation showed the analogy to be false, for these amylases possess properties unlike those observed in any previous investigation of the cereal amylases. The present paper reports a study of the insoluble amylases of birdproof kaffircorn malts and a comparison of their behaviour with those of barley malts.

It is well known that only a small proportion of the β-amylase of wheat and barley is soluble and that germination causes a conversion of the insoluble amylase to a soluble form. The insoluble amylase may be partially or completely extracted by a variety of reagents such as salt solutions, proteinases and reducing agents. The nature of the insoluble amylase and the extraction process has been the subject of considerable investigation and controversy.

Three fractions of barley β-amylase may be distinguished: (*a*) water-soluble (' free ' or ' active ' amylase), (*b*) salt-soluble and (*c*) that brought into solution by the action of proteinases or reducing agents. The latter two fractions are inactive in the malt but become active when brought into solution.

The inactivity of the salt-soluble amylase is regarded as being due to physical causes. Dull & Swanson[2] postulate that the enzyme is adsorbed on, or enveloped in, a protein film which prevents its action. In other words the active centre of the amylase is fully functional but its action is prevented by a physical barrier. Salt solutions peptise the enveloping protein and thus free the amylase. Erlich & Burkert[3] and Pollock & Pool[4] do not regard the water-soluble and salt-soluble amylases as being fundamentally different. While this may well be so, it is useful practically to distinguish between the two.

Salt solutions do not bring all the insoluble amylase into solution ; there is a portion which is rendered soluble only by means of proteinases or reducing agents. The insolubility and inactivity of this particular fraction is believed to be due to its being attached to another protein and when the link is broken, the enzyme becomes both soluble and active. There have been three theories as to the nature of this enzyme–protein compound and the manner in which the amylase is liberated. According to Myrbäck[5] the liberation of insoluble amylase is a proteolytic process which may take place in one of three ways :

(i) the proteinase of the grain may, if allowed to act for a long enough period during aqueous extraction, degrade the protein binding the amylase.

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† Kaffircorn Diastatic Units (see reference [1], p. 444)

(ii) Hydrogen sulphide or cyanide may be added to accelerate the action of the naturally occurring proteinases thus giving a more rapid or more complete release of amylase.

(iii) The action of the natural proteinases may be strongly supplemented by the addition of a proteinase such as papain.

The third method of release gives the highest yield of amylase ; the others generally give lower values depending on how active the natural proteinases are. Myrbäck recognises only two fractions, active, water-soluble amylase and inactive insoluble amylase. The theory therefore offers no explanation of the nature of the salt-soluble amylase. Chrzaszcz & Janicki[6] and Sandegren & Klang[7] have questioned the proteolytic theory on the grounds that no parallel could be found between the amount of amylase liberated by the papain and the increase in soluble nitrogen resulting from the protein degradation. Myrbäck[5c, 8] stated that such a parallel was not to be expected because the amylase was most certainly bound, not to all the proteins present, but to one particular protein whose degradation contributed only a negligible amount of nitrogen to the total quantity produced by the proteolysis. Such a claim is entirely reasonable. According to the proteolytic theory, the effect of cysteine and similar reducing agents in liberating amylase would be due to their ability to stimulate the natural proteinases of the grain or malt.

Chrzaszcz & Janicki[9] put forward an alternative theory to explain the action of proteinases and reducing agents. They considered that the β-amylase of cereals was held insoluble and inactive by ' sisto ' (inhibitor) substances. The enzyme–inhibitor complex could be dissociated by the action of peptone or of hydrogen sulphide. Proteinases were only indirectly effective ; the products of their action, resembling peptone, were the direct cause of the liberation of amylase. A long controversy between these workers and Myrbäck as to the validity of their respective methods of extraction and estimation was not settled until 1939/40 when Myrbäck & Örtenblad[8, 10], cf.,[11] showed that the Polish workers' results were erroneous because of the presence of copper in the distilled water used, copper being a powerful inhibitor of amylases. Hydrogen sulphide reversed the inhibition caused by the copper and peptone was regarded as functioning in a similar manner.[8] [These papers of Myrbäck & Örtenblad appear to have been overlooked by a number of workers who still include the theory of Chrzaszcz & Janicki in their reviews.]

The use of peptone in the liberation of amylase from the common cereals is of interest to the present work where peptone has been found to be an excellent extractant for the insoluble amylases of kaffircorn malt under circumstances where copper inhibition is excluded. Myrbäck[5b] showed that peptone extracted no more amylase from barley than did water. Hills & Bailey[12] confirmed this, but found with malts that the addition of peptone to already extracted α-amylase caused activation of the enzyme. Kneen *et al.*[13] found that peptone extracted more α-amylase from wheat than did water but less than that released by papain. No activation of α-amylase already extracted was found. Further, Ford & Guthrie[14] observed that boiled papain extracted more amylase from barley than did water but never as much as active papain. On the other hand Myrbäck[5b] found that inactive papain was without action on bound β-amylase. While it is reasonable to explain the effect of peptone as a reversal of inhibition in the case of the Polish investigators, this is unlikely to have been the explanation in all other instances quoted.

A third theory postulates that release of the amylase is due to reductive scission of a disulphide bond, although no clear and detailed formulation of this theory is available in the literature. It appears to have been developed by Sandegren & Klang[7] from a suggestion of Erlich & Burkert[3] based on the work of Weill & Caldwell[15] on purified soluble β-amylase in which was demonstrated the presence of a sulphydryl group essential to its activity. Oxidation to the disulphide caused inactivation which could be reversed by reduction with hydrogen sulphide. Erlich & Burkert postulated a similar reductive reaction when insoluble β-amylase was brought into solution by papain. One may envisage the following reaction as taking place with cysteine :

$$\text{Enzyme-S-S-protein} \xrightarrow[\text{papain}]{\text{cysteine or}} \text{enzyme-SH} + \text{protein}$$

Both this theory and the proteolytic theory fit the available facts equally well and it is not possible at this stage to decide in favour of either.

The malting of barley, wheat and rye results in the solubilisation of the inactive fraction of the β-amylase.[13, 16] According to Myrbäck,[16] incomplete conversion to the soluble form is the result of insufficient modification of the malt. Kneen and co-workers[13] find accelerated conversion of insoluble wheat β-amylase to the soluble form at higher temperatures, for instance complete conversion was reached in 4 days at 20° instead of the 10 days necessary at 10°. Erlich & Burkert[17] report that wheat malts contain relatively large proportions of salt-soluble β-amylase but barley malts very little.

The α-amylase which is formed during germination exists largely in the soluble form throughout malting. According to Kneen,[13, 18] 80–90% or more of the total α-amylase is soluble; peptone releases more α-amylase from wheat malt than does water but considerably less than do papain extracts.[13] No evidence was presented as to whether the α-amylase in its insoluble form was active or not. Considerable quantities of salt-soluble α-amylase have been found in both brewers' and distillers' malts,[17, 19, 20] but papain is a less efficient extractant of barley malt α-amylase than salt solutions.[19] Lowry & Olson[20] found the quantity of α-amylase extracted by water to vary with the concentration of the malt used in the extraction. This was probably due to an increase in the concentration of salt which is naturally present in the malt. In this connexion it has been noted[3, 14] that the natural and variable salt content of grain must have an effect on the aqueous extraction, so that the proportion of salt-soluble amylase can be expected to vary from sample to sample and with the concentration used in extraction.

From this brief review, it is clear that although the state of the amylases in barley and the changes which occur on malting have been studied intensively, there is no general agreement as to what occurs. For this reason certain aspects of the extraction of the amylase from barley and barley malt have been investigated in parallel with those of the kaffircorn malt amylases.

Experimental

Extraction and estimation of amylase

The extraction and estimation were carried out essentially as described in the preceding paper of this series.[1]

Extracting agents.—These were of the highest purity available and were tested to see if they inhibited amylase activity. Where necessary the pH of the solution was adjusted to 6–7 before extraction. Only freshly neutralised solutions of cysteine hydrochloride were employed for extraction. The papain used had no amylase activity and essentially the same results were obtained using B.P.C., Hopkin & Williams, or Merck's preparations.

Time of extraction.—Unless otherwise stated, extractions were carried out for 2½ h. at 30°. The period of extraction was taken as ending when filtration was started, except in the case of very brief extraction periods (1 h.) when the times include the time for filtration. The period required for filtration was cut to 2 min. or less by filtering by suction the minimum volume required for the estimation and by using a large Buchner funnel for rapid filtration.

Extraction of undried kaffircorn grain.—The grain samples were crushed in a mortar since they could not be milled. After being crushed, the grain was ground with successive portions of the extracting solution and the suspension formed transferred to the extraction flask. Extraction was then carried out as usual.

Extraction pH and diastasis.—The pH of the extracts was always adjusted to 5–6 prior to diastasis so as to maintain the optimum for amylolysis.

Malts

The malts were made in the laboratory by a technique specially developed for kaffircorn to give well-modified malts of high diastatic power. The details and development of this malting procedure will be the subject of a further communication. In brief, the grain was steeped for 8–18 h. in running water and germinated at the optimum temperature for amylase development, 30°, for up to 7 days (previous investigators have used either too low a temperature[21] or too short a germination period[18a, 22]). The malt was dried in a forced-draught oven at 50°, which temperature has been proved to cause no destruction of amylase.

Results and discussion

I. *Extraction of the amylase of birdproof kaffircorn malt*

(a) *Papain*

By analogy with barley, papain was tried as an extractant for the insoluble amylases of the kaffircorn malts. It proved capable of liberating considerable quantities of amylase from birdproof kaffircorn malts; some typical results are given in Table I. The relatively high concentrations of papain required for maximum liberation of amylase were not reduced by the use of more active preparations, nor by the addition of an activator.

Inactive (boiled) papain solutions were used for extraction to see whether proteolytic action was essential. As the results in Table II show, inactive papain is fully as capable of liberating the amylases from birdproof malts as is the active preparation. This is in complete contrast to Myrbäck's findings.[5b]

Table I

Extraction of birdproof kaffircorn malts with papain

(5 g. of malt extracted with 100 ml. of papain solution for 18 h. at 30°)

Malt no.	Papain %	Amylase activity extracted (K.D.U./g.)
M54/18	0	0·3
	0·5	18·0
	2·0	35·2
	5·0	39·8
	10·0	38·4
M54/19	0	0
	2	37·0
M54/33	0	0
	10	28·3
O.M.16/4	0	4·1
	5	49·3

Table II

Extraction of birdproof kaffircorn malt with active and inactive papain

(5 g. of malt M55/11 extracted with 100 ml. of papain solution for 18 h. at 30°)

Papain concn., %	Treatment of papain solution used for extraction	Amylase activity released (K.D.U./g.)
0	Usual aqueous extraction	1·7
5	None	38·0
5	Boiled	37·1
5	Autoclaved	32·9
5	Dialysed, buffered to pH 6	34·1
5	Dialysed, boiled and buffered to pH 6	35·3
5	Dialysed, boiled and buffered to pH 6, cysteine added (4%)	40·2

The liberating action of inactive papain may be due to (i) the presence of impurities in the papain preparation; (ii) its general properties as a protein; (iii) some special group or groups in the protein.

The usual commercially available preparations of papain are known to contain activators (sulphydryl compounds) of low molecular weight as well as salt as impurities. Dialysis of active or inactive papain solutions produced no effect, however, on the efficacy of the preparation (Table II), so that the liberation of amylase is not due to the presence of impurities of low molecular weight in the papain. However, the papain molecule itself is known to contain a sulphydryl group and the release of amylase could possibly be due to this and, to clarify this point, the effect of other proteins and of sulphydryl compounds was studied.

(b) *Proteins and peptone*

Casein, egg albumin and peptone proved to be excellent extractants, liberating as much amylase as did papain (Table III). Hydrolysis of the proteins caused a reduction in the quantity of amylase liberated.

Peptone ('Bactopeptone') was chosen for further study since it is a standard product, readily soluble and free from sulphydryl groups (nitroprusside test). Table IV shows that a concentration of 2% peptone is sufficient for maximum extraction of amylases. Tests showed that Bactopeptone used at its normal pH (6·5) gives satisfactory extraction.

Time of extraction.—Although an extraction period of 2½ h. is required for quantitative extraction, the greater part of the insoluble amylase passed into solution with remarkable rapidity (Table V). Where a sub-optimal concentration of peptone (0·1%) (Table IV) was used, extension of the time of extraction yielded no increase in the quantity of amylase extracted.

Table III

Extraction of birdproof kaffircorn malt with proteins and protein hydrolysates

[Malt 55/11 (5 g.) extracted with 100 ml. of extractant (2%)]

Extractant	Amylase activity released, K.D.U./g.
Peptone	38·5
Egg albumin	39·1
Casein	38·4
Peptone hydrolysate	25·8
Egg albumin hydrolysate	28·1

Table IV

Effect of concentration of peptone on extraction of birdproof kaffircorn malt amylases

[Malt (5 g.) extracted with peptone solution (100 ml.) for 2½ h. at 30°]

Peptone concn., %	Amylase activity extracted, K.D.U./g., from malts		
	M55/11	M55/50	M57/24
0·1	20·1	—	—
0·5	32·8	—	—
1·0	37·3	38·4	—
2·0	36·8	41·5	48·0
5·0	36·5	41·1	47·1
10·0	—	40·2	—

Table V

Rapidity of peptone extraction of amylases of birdproof kaffircorn malts

[Malt (5 g.) extracted with peptone solution (2%, 100 ml.) for various times with stirring or in Waring Blendor]

Malt No.	Method of extraction	Time of extraction	Amylase activity released, K.D.U./g.
M55/14	Continuous stirring	15 min.	35·2
		1 h.	35·9
		3 h.	35·4
		5 h.	40·2
		24 h.	39·6
	Waring Blendor	6 min.	36·5
M57/24	Occasional stirring	2½ h.	48·7
	Continuous stirring	4 min.	43·7

Since peptone and casein have no enzyme activity, their effectiveness in liberating the amylases of birdproof kaffircorn malts cannot be attributed to direct proteolytic action. Further, since peptone has no sulphydryl groups, stimulation of the natural proteinases of the malt to effect release of the amylases is clearly impossible. The rapidity of release of the amylases by peptone also argues against an enzymic mechanism of release. The data are consistent with the liberation being a physical desorption of amylase effected by a variety of proteins.

If the effectiveness of the protein lay solely in the presence of peptide linkages or in some general polymer-type of property, it would be expected that hydrolysis would cause complete loss of extractive ability. If, on the other hand, the effect were due solely to a particular amino-acid or to a number of amino-acids acting individually, no reduction in extractive capacity should be found on hydrolysis. The results in Table III indicate that both effects are found, i.e., liberation of amylase is due both to protein (polymer) properties and to amino-acids.

To study these aspects of amylase release, the effect of surface-active agents and amino-acids was investigated.

(c) *Surface-active agents*

Only the non-ionic reagents proved to be good extractants for the insoluble amylases, their effectiveness increasing with chain length particularly in the range $n = 1$ to $n = 10$ (Table VI). These compounds generally released slightly less amylase than did 2% peptone.

(d) *Amino-acids*

Since the extractive capabilities of peptone could be due, in part at least, to the presence of certain amino-acids in the molecule, a number of amino-acids were examined for their ability to extract amylases (for cysteine, see below).

The basic amino-acids were the most effective, liberating 15–45 K.D.U./g., while all others liberated less than 10 K.D.U./g. of malt 55/11 (see Table VI for aqueous and peptone values).

Table VI

Extraction of the amylases of birdproof kaffircorn malts with surface-active agents

[Malt (5 g.) extracted with aqueous solution of surface-active agent (100 ml.)]

Extractant	Concentration, %	Amylase activity extracted, K.D.U./g., from malt		
		55/11	57/45	L84IVD6
(*a*) $C_8H_{17}\cdot C_6H_4\cdot(OCH_2\cdot CH_2)_n\cdot OH$				
$n = 1$* (Triton X-15)	0·5	—	7·5	—
$n = 5$ (Triton X-45)	0·5	—	19·0	32·1
$n = 9$–10 (Triton X-100)	0·05	24·0	—	—
	0·1	30·1	—	—
	0·2	32·7	—	—
	0·5	35·7	26·9	44·1
$n = 12$–13 (Triton X-102)	0·5	—	29·2	46·7
$n = 16$ (Triton X-165)	0·5	36·6	28·1	49·8
$n = 20$ (Triton X-205)	0·5	32·4	29·2	50·3
(*b*) Water	—	1·7	4·6	5·8
(*c*) Peptone	2	40·4	29·2	53·2

* Partly as an emulsion because of low solubility

The order of effectiveness of the basic amino-acids on an equimolar basis is histidine > arginine > lysine, which is not in order of their basicity.

With the exception of histidine, no amino-acid liberated more amylase than did peptone. The concentrations of amino-acids required for complete or near-complete liberation of amylase were far in excess of the concentrations in which they could occur in a 2% peptone solution.

(e) *Amines and related compounds*

The efficacy of the basic amino-acids could be due to their basic nature or to some characteristic common to their molecules, e.g., to the presence of two or more nitrogen atoms. The fact that histidine, the least basic of the three, is the most effective in extracting the insoluble amylases would seem to lend support to some alternative to the need for a basic substance for extraction, but the possibility that a certain specific degree of basicity might be required could not be entirely dismissed. For these reasons the extractive action of a number of amines and related compounds were studied.

Of mono-, di- and triethylamine, mono-, di and triethanolamine, tetraethylammonium chloride, tris(hydroxymethyl)aminomethane and ethylenediamine, the most effective compound was the diamine, the next, triethanolamine. The effectiveness of the amines in liberating amylase was closely comparable with that of the basic amino-acids. As with the amino-acids, basicity and effectiveness did not go parallel to one another.

With two exceptions (histidine and ethylenediamine) all the compounds tried at this stage did not release as much amylase as did peptone, but this was shown not to be due to their possessing any inhibitory action on amylolysis. It follows therefore that the presence of a basic group in the molecule of liberating agent is only a part of the requirements for complete liberation. The most effective extractants, histidine and ethylenediamine, contain two or more nitrogen atoms to the molecule. Examination of a number of polynitrogenous compounds, especially purines and pyrimidines, showed that these compounds could be divided into two groups: those that extracted less than 25% and those that extracted 80% or more of the amylase extracted by peptone. The low values obtained with the first group of extractants were not due to their inhibiting amylase activity: these included alloxan, allantoin, biuret, creatine, hydantoin, uracil and uric acid. In the second group were adenine, 2-aminopyridine, 2-aminopyrimidine, caffeine, creatinine, hexamine, theophylline and 4,5,6- triaminopyrimidine. A comparison of the structure of these compounds (and of arginine and histidine) showed that all effective compounds contained the group—N·C(X)·N—, where X is not oxygen. This group is effective whether it is part of a ring or of a side chain. Compounds containing an —N·CO·N— group only, whether it is in a ring or chain, are ineffective. The group —N·C(X)·N— may fail to confer ability to extract amylases if it is flanked by CO groups as in allantoin and uric acid. It is possible, however, that these two substances are ineffective because of their low solubility.

Another interesting exception was creatine which, in spite of its structural resemblance to arginine, did not liberate amylase. It was concluded from this that the ineffectiveness was the result of the close proximity of a negatively charged carboxyl group to the guanidyl group, since in arginine these groups are well separated. This was strikingly confirmed by the effectiveness of creatinine in extracting amylases, where the charge has been lost by the conversion to the cyclic amide.

(f) *Salts*

In view of the existence of a salt-soluble amylase fraction in barley, the effect of salts on the extraction of the sorghum amylases was investigated.

Sodium chloride solutions extracted more amylase from birdproof kaffircorn malts than did water. Maximum extraction was obtained with 2·0–2·5M-sodium chloride solutions. The values obtained, however, were always lower than with peptone extraction (Table VII). As in the case of barley therefore, part, but not all, of the insoluble amylase is extracted by salt solutions. The concentration of salt needed to achieve maximum extraction is much higher however for birdproof kaffircorn malts than for barley (cf. Table XII), although maximum extraction is obtained from the normal type of kaffircorn malt with water alone (Table VII).

Table VII

Extraction of birdproof kaffircorn malts with salts

Extractant and molarity	Amylase activity extracted, K.D.U./g.			
	Birdproof kaffircorn malts			Non-birdproof kaffircorn malt
	M54/19	M55/11	M55/50	
Peptone (2%)	37·0	40·4	41·0	—
Water	0	1·7	0	29·1
Sodium chloride				
0·1	0	11·2	2·3	28·7
0·5	5·1	20·5	10·9	—
1·0	8·9	24·9	16·3	28·3
2·0	11·6	35·4	33·0	25·9
2·5	25·2	29·2	—	—
3·0	25·3	31·3	32·5	23·1
4·0	19·5	29·8	30·5	—

The fact that the insoluble kaffircorn malt amylases are extracted by salt concentrations of the same order as those commonly used to extract globulins and nucleoproteins suggests that the amylases liberated by the salt may either belong to these classes of proteins or be associated with them. The latter hypothesis seems the most likely since the amylases are not precipitated along with the globulins and nucleoproteins when a sodium chloride extract of birdproof kaffircorn malt is dialysed, i.e., once extracted, the amylases remain soluble in water. It is also supported by the fact that the amylases are liberated by peptone and surface-active agents which can be visualised as causing the desorption of the enzymes from the surface of the protein to which they are bound. The rapidity with which the amylases are brought into solution by the peptone and the fact that they are active in the insoluble or adsorbed state suggests that the enzyme molecules lie wholly on the surface of the adsorbing material and that the active centres are not involved in the binding. This contrasts with the case of the salt-soluble β-amylase of wheat and barley which is supposed to be enclosed by a globulin in such a way as to prevent the active centre of the enzyme from acting on starch.[2]

(g) *Ribonuclease*

Because a nucleoprotein might be the adsorbent for the insoluble amylases, the effectiveness of ribonuclease as an extractant was examined. Ribonuclease released as much amylase as an equivalent quantity of peptone (Table VIII). Extension of the extraction time from 2½ to 6 h. did not increase the yield of amylase. These observations, together with the fact that inactivated ribonuclease also liberated amylase (Table VIII), indicate a non-enzymic mechanism of release similar to that observed with casein, peptone, etc. Although this is not conclusive

evidence against the hypothesis that the amylase is adsorbed on a nucleoprotein, it gives it no support. It therefore appears most likely that the substance to which the amylase is bound is a globulin.

(h) *Acid and alkali*

The ability of the organic bases to effect partial liberation of the adsorbed amylases prompted the trial of sodium hydroxide as a possible extractant. The extraction was accomplished by titrating a chilled suspension of the malt in water with sodium hydroxide until the pH rose to the required value within the range 7–9·5. After being kept for 5, 15 or 60 min., the mixture was filtered and the activity of the filtrate determined after adjustment of the pH to 6.

Maximum extraction of amylase was obtained at pH 9. The extraction although rapid (90% of the maximum yield from malt 55/11 being obtained in the first 5 min.) was, however, far from complete, little more than half the peptone-extractable amylase being released. The action of sodium hydroxide does not appear to be a salt effect since the final molarity of the alkali in the mixture was less than one-twentieth of the sodium chloride concentration required to release an equal quantity of amylase (cf. Table VII).

A birdproof kaffircorn malt with a higher proportion of water-soluble amylase than the M55/11 malt was extracted with acetic acid by a procedure similar to that used with the alkali. At pH 4, only 0·7 K.D.U./g. was released in comparison with the 18·1 K.D.U./g. obtained with aqueous extraction, and the 69·0 K.D.U./g. with peptone. Acid, therefore, caused a retention of amylase. The results suggest that the insoluble amylases are held, at least in part, by the charge differences between them and the adsorbing material, and that the reduction of the electropositive character of the amylase by the addition of alkali permits extraction to take place.

Table VIII

Extraction of birdproof kaffircorn malt with crystalline ribonuclease

(malt L79IID4)

Extractant	Amylase activity extracted, K.D.U./g.
0·1% ribonuclease	26·4
0·1% inactivated ribonuclease	25·0
0·1% peptone	26·5
2% peptone	52·2
water	8·4

(i) *Cysteine*

Determinations of diastatic power by the direct method[1] have shown the greater part of the insoluble amylase to be active. Nevertheless, it was possible that a small amount of β-amylase was present in the malt in an insoluble and inactive form. Because of the effectiveness of cysteine in liberating the bound, inactive β-amylase of barley,[7] the extraction of birdproof kaffircorn malts was tried with varying concentrations of cysteine.

The quantity of amylase liberated increased in an unexpected manner with the concentration of cysteine (Table IX), since normally, only low concentrations are needed for maximum liberation of amylase from barley.[7] All cysteine hydrochloride solutions used in the present work were neutralised with sodium hydroxide before use; by calculation of the quantity of sodium chloride thus formed and reference to Table VII it was shown that the greater part of the amylase released by the cysteine solution could be attributed to the salt present. To eliminate the salt effect, a series of extractions were made with cysteine in conjunction with sodium chloride and peptone. Addition of cysteine to 2·5M-sodium chloride led to a slight increase in the amount of amylase extracted (Table IX). This cannot be due to the additional salt formed by the neutralisation of the cysteine hydrochloride because 2·5M-sodium chloride is already above optimum salt concentration for maximum release of the amylases (cf. Table VII). Addition of cysteine to a 2% peptone solution gave a similar increase in amylase released. Again this cannot be attributed to a salt effect because M-sodium chloride does not increase the effectiveness of aqueous peptone. Therefore, it may be concluded that cysteine releases a small amount of insoluble inactive amylase in birdproof kaffircorn malt which is not extracted by other means.

The addition of cysteine (0·1%) to a filtered peptone extract of malt produced no increase in activity (Table IX), so that the inactive amylase is insoluble. This was substantiated by the

Table IX

Extraction of amylases of birdproof kaffircorn malt with cysteine

[Malt M55/11 (5 g.) extracted with reagent (100 ml.). pH adjusted to 6–7 where necessary]

Extractant		Amylase activity extracted, K.D.U./g.
Cysteine hydrochloride	0·3%	7·3
	2·0%	16·0
	4·0%	17·7
Peptone 2% + cysteine	0%*	37·7
	0·1%	41·3
	0·5%	45·4
	1·0%	45·4
* Filtrate from this extraction + cysteine		38·4

Extractant		Amylase activity extracted, K.D.U./g.
Sodium chloride 2·5M + cysteine	0%	23·1
	0·12%	26·4
	2·0%	27·8
	4·0%	29·5
Peptone 2% in M-sodium chloride solution		38·0

fact that re-extraction of the malt residue with the cysteine and peptone filtrate gave a filtrate with the same activity as was obtained when the malt was extracted directly with peptone and 0·1% cysteine.

(j) *Effect of water on the extraction of insoluble amylases*

As has already been noted in the description of the direct diastatic power and direct dextrinisation methods,[1] the addition of water to the malt prior to its addition to the starch decreased its activity significantly. In one instance, the apparent diastatic power of the malt fell from 25·5 to 12·8 K.D.U./g. and the α-amylase activity from 48·5 to 30·0 30°-dextrinisation units. Although the time of contact between malt and water before their addition to the starch solution was very brief (a matter of seconds), this was sufficient to reduce the saccharification activity by approximately 50%.

Table X

Effect of water on the state of the birdproof kaffircorn malt amylases

[Four portions of malt M55/11, 5 g. each, extracted with water (100 ml.) for 0·5, 2, 3 and 48 h.; peptone (2 g./100 ml.) added to all except the 48 h. extract; mixtures filtered after 20 min.]

Duration of aqueous extraction before peptone was added, h.	Amylase activity released, K.D.U./g.
0·5	5·6
2·0	6·1
3·0	6·5
48·0*	3·2
(Peptone extraction)	(40·0)

* No peptone added

Further investigation (Table X) showed that contact with water for as short a period as 30 min. rendered the amylases almost completely non-extractable by peptone, while prolonged (48 h.) extraction with water gave no increase in the amount of amylase extracted. This contrasts with the bound amylase of barley where prolonged aqueous extraction results in a considerable increase in the amount of amylase extracted.[3, 8]

Water thus has two effects on the insoluble amylases of birdproof kaffircorn: it prevents their reaction with starch and also their subsequent extraction with peptone. At present no explanation of this phenomenon can be offered. It is interesting, however, to compare the remarkable action of water in these instances with the curious effect it has on the dormancy of barley.[23]

II. *Comparative studies on extraction of the amylases of barley and barley malt*

Because of conflicting statements in the literature about the insoluble amylases of the common cereals, it was difficult to compare them with the insoluble amylases of kaffircorn malts. Samples of barley and barley malts were therefore analysed under the same conditions as were used for kaffircorn malts. The results are given in Tables XI and XII. Whereas papain and peptone were equally effective in the case of birdproof kaffircorn malt (cf. Tables II and III), for barley, papain was considerably more effective than peptone, which did not extract much more amylase than water (Table XI). Contrary to the findings of Myrbäck,[5b] peptone exerts a definite but minor effect, assisting the release of a very small quantity of amylase which is

apparently held more firmly than the remainder of the water-soluble enzyme. With the malts, the peptone appeared to speed up the release of amylase in the early stages, and also to give a slightly higher final yield of amylase.

Cysteine was an excellent extractant for both barley and the malts. The speed of action of cysteine is noteworthy. In only 6 min. cysteine extracted from barley three times the quantity of amylase extracted by water in 15 min., and more than half the total amylase of the grain (Table XI). This contrasts strongly with the very limited effect of cysteine on birdproof kaffircorn malts over a 2½-h. period (Table IX). The activity extracted from barley and

Table XI

Extraction of the amylases of barley and barley malt

Material	Extraction time, h.	Amlyase activity extracted			
		Water	Peptone 2%	Papain 2%	Cysteine 1%
Barley 1/1	0*	29·6*			
	0·15				71·7
	0·2	24·2	30·0	88·3	99·1
	2·5	52·0	60·1	124·2	120·6
	18·0	71·3	75·8	135·0	125·5
Malt 1/1	0*	71·0*			
	0·1				100·7
	0·25	68·9	88·9	103·1	113·2
	2·5	92·8	95·8	110·1	122·7
	18·0	92·8	98·8	118·4	110·5
Malt 2/1	0*	52·9*			
	0·1				73·1
	0·25	49·9	62·6	74·4	79·3
	2·5	64·4	69·6	75·3	84·9
	18·0	61·3	76·2	78·8	76·6

* Direct diastatic power estimations, i.e., no extraction

Table XII

Extraction of barley and barley malts with salt solutions

Molarity of extracting sodium chloride	Amylase activity extracted, K.D.U./g. from		
	Barley 1/2	Malt 1/2	Malt 2/1
0	56·2	74·7	52·3
0·1	59·9	79·1	64·7
0·5	61·7	81·3	66·9
1·0	57·6	81·3	62·4
2·0	50·7	76·9	62·0
3·0	45·7	76·0	60·6

malt by water in 15 min. was very close to that found in the direct diastatic power determination. It is obvious therefore that only the water-soluble portion of the total amylase is active in the direct diastasis. This is in marked contrast to the behaviour of birdproof kaffircorn malts where the diastatic power determined by the direct method is always far higher than that found by aqueous extraction. The insoluble amylases of barley and of birdproof kaffircorn malt are thus entirely different in their behaviour during direct diastasis.

Maximum yield of amylase from barley and barley malt was obtained by extraction with 0·5M-sodium chloride (Table XII), whereas the conditions of assay were such that the concentrations of sodium chloride during amylolysis could not exceed M/70. The lower amylase values obtained with the higher salt concentrations could not therefore be due to inhibition of amylolysis, as claimed by Dull & Swanson.[2] The lower values were probably due to precipitation, either of the enzymes themselves or of other proteins with subsequent adsorption of the enzymes on them. In some cases, actual coagulation of a portion of the ground material was clearly seen to take place during the extraction. As pointed out previously, the salt concentration which gives the maximum extraction of the barley enzymes is very much lower than that required in the case of kaffircorn malt.

III. *Occurrence of amylase inhibitors in kaffircorn*

Kneen and co-workers have reported the presence of amylase inhibitors in wheat and sorghum (kaffircorn);[24] consequently, some South African sorghum grains were investigated to see if the presence of such an inhibitor could explain the anomalous behaviour of the amylases of birdproof kaffircorn malts.

Aqueous extracts of birdproof kaffircorn grain were found to contain a weak inhibitor which when added to malt extract reduced the amylase activity by 9–14%. A still smaller degree of inhibition was found with extracts of short red kaffircorn, but no inhibitor was found in the malts of birdproof or short red kaffircorn. The low amylase activity of aqueous extracts of birdproof kaffircorn malts could not therefore be due to inhibition and even if a large quantity of unmalted grain were present in the malt (which is very unlikely), it could not cause a sufficient depression of activity to account for the low values found.

IV. *Factors influencing the occurrence of insoluble amylase in kaffircorn malts*

(a) *Absence of amylase from all varieties of kaffircorn grain*

It has been established for wheat (Schwimmer[25]) and for rye (Chrzaszcz & Janicki[26]) that in the early stages of the development of the grain in the ear, the amylase is soluble, but becomes insoluble as ripening progresses. Few studies have been made on ripe kaffircorn and none on the immature grain. Of the few American varieties examined, all have been shown (using water or papain for extraction) to contain very little of either α- or β-amylase.[18a, 27] In view of the difference in β-amylase content of South African and American varieties of kaffircorn malt and the exceptional properties of birdproof kaffircorn, it was decided to examine both mature and immature South African kaffircorns for amylase activity by extraction with water and papain and also with peptone.

With none of the three extractants was it possible to extract more than 0·4 K.D.U./g. from any of the mature kaffircorn varieties examined, viz., short red (2 samples), birdproof (3 samples) and white. In order to determine the traces of amylase activity present in the extracts of the immature grain, the period of diastasis was extended from $\frac{1}{2}$ h. to 4 h. In spite of this, the values found were significant in only two cases out of five (Table XIII). Therefore, kaffircorn, whether of a normal or birdproof type, whether mature or immature, contains no more than the merest traces of amylase activity. Consequently, germination must result in the formation of both α- and β-amylase, either in a soluble or in an insoluble form.

Table XIII

Amylase activity of immature birdproof kaffircorn

Stage of growth of grain	Extraction	Diastatic power, K.D.U./g.
Milk stage, undried	2% papain, 18 h.	0·12
Early dough stage, undried	2% peptone, 18 h.	0·2
Early dough stage, air-dry	2% peptone, 18 h.	0·0
Late dough stage, undried	Water, 2½ h.	0·01
	Peptone, 18 h.	0·03

(b) *Variety*

The greater part of the kaffircorn malt of commerce is made from the so-called K2 red kaffircorn, which is not a pure variety but includes a number of reddish coloured, probably closely related, varieties. None of these commercial malts contains significant amounts of insoluble amylase. Insoluble amylases were first found in malts made from the birdproof variety of kaffircorn, use of which raised certain analytical difficulties (see Part III of this series)[1] and also questions as to its suitability for brewing. It became important, therefore, to see whether there were not other varieties of kaffircorn whose malts possessed similar characteristics.

From Table XIV, it is seen that the amylases of malts made from the common varieties of grain sorghum, viz., short red (a variety typical of those comprising the K2 red grade), Martin, Hegari and Radar are almost entirely soluble. These varieties will be referred to as 'normal' or 'non-birdproof' types. Besides birdproof, only two other varieties of grain sorghum—

Table XIV

Ratio of soluble to total amylase in different types of kaffircorn malt

Variety	Days of germination	Total* amylase, K.D.U./g.	Soluble/total amylase, %
Grain sorghums			
Short red	2	29·7	89·5
	3	56·8	99·1
	4	66·4	95·9
	5	68·5	100·0
	6	64·3	95·6
Martin	4	60·0	84·0
	5	64·3	95·3
	6	65·8	101·7
Swazi red	5	70·3	78·3
Hegari	5	55·9	90·3
	6	45·7	96·5
	7	44·0	93·9
Radar	4	65·5	93·7
	5	66·3	91·0
Framida	6	64·2	55·8
Sweet sorghums			
Haakdoring	7	72·9	47·2
Sugar drip	7	56·5	19·3
Black hull	7	32·4	0

* Peptone extraction

Swazi red and Framida—have so far been found to give malts which contain an appreciable proportion of insoluble amylase, though rather less than in the case of birdproof kaffircorn (Table XIV).

Little is known about the genetical relationships of these different varieties of sorghum. Birdproof kaffircorn is regarded by agricultural authorities,[28] on morphological grounds, as being a cross between a grain sorghum and a sweet sorghum (*Sorghum saccharatum*). A selection of sweet sorghum varieties yielded malts containing a high proportion of insoluble amylases. The amylases of the 'black hull' variety were actually completely insoluble in water (Table XIV). The relationship between the sweet sorghums and birdproof kaffircorn is thus confirmed by the similarity in the properties of their amylases.

Of the two varieties of grain sorghum which yielded insoluble amylases, Swazi red is regarded as being a cross between a typical grain sorghum and birdproof kaffircorn. Here again there is confirmation of relationship from the properties of the malt amylases. The other variety, Framida, a grain from French Equatorial Africa, has a superficial resemblance in colour and shape to birdproof kaffircorn, but lacks the dark nucellar layer which characterises the latter and the sweet sorghums. In this case the evidence provided by the amylases suggests a possible genetical relationship which may be worth investigation.

(c) *Effect of conditions under which the malt is made*

(i) *Temperature and time of storage of grain before malting.*—Birdproof grain malted soon after harvest gives malts with a low soluble amylase content (Table XV), but after 2–3 years' storage at room temperature (12–33°), the grain furnished malts with a considerably higher

Table XV

Ratio of soluble to total amylase in birdproof kaffircorn malts

Grain	Days of germination	Total amylase, K.D.U./g.	Soluble/total amylase, %
Malted 28.9.54 about 5 months after harvest	4	49·3	8·3
	5½	49·8	22·1
	6	53·6	15·5
	7	47·0	24·0
Stored at room temp., malted 17.12.56	6	62·1	57·5
Stored at 7°, malted 10.1.57	4	59·8	16·1
	5	67·0	20·0
	6	74·0	30·5
Stored at room temp., malted 5.9.57	3	57·2	61·0
	4	66·6	63·5
	5	78·4	69·8
	6	85·2	71·2
	7	85·6	72·7

proportion of soluble amylase, viz., between 57 and 73% of the total as compared with under 25% for malts from fresh grain. Grain stored for about 3 years at 7° showed little change in its malting characteristics, the soluble amylase being less than 31% of the total amylase. At room temperature the birdproof kaffircorn thus appears to undergo a very slow maturing process, as a result of which malts made from it have an increased content of soluble amylase, although storage of the malt has practically no effect on the soluble amylase content.

(ii) *Duration of malting.*—The proportion of soluble to total amylase increases slightly during malting with both the freshly-harvested and the stored grain. In no case did complete solubilisation result even in the well-modified malts of 7 days' germination (Table XV).

General conclusions

The present studies have shown that kaffircorn grain has no more than traces of soluble and insoluble amylases. Malting of the grain results in the formation of both α- and β-amylases which may be either completely soluble or very largely insoluble according to the variety of the grain. The insolubility of the amylases has been found in malts made from the sweet sorghums and related varieties, particularly birdproof kaffircorn. Prolonged storage of this variety results in the malts made from it having a greatly increased content of soluble amylase but complete solubilisation of the amylases is not attained.

This is in marked contrast to the behaviour of barley and wheat, where the bound portion of the β-amylase present in the ungerminated grain becomes soluble on germination. Further, the α-amylase produced at the same time is largely soluble,[13, 18] whereas in birdproof kaffircorn malts the α-amylase is largely insoluble. The insoluble amylases of the latter malts are active, whereas that of barley is inactive.

With birdproof kaffircorn malts, prolonging the time of aqueous extraction gives no increase in the amylase extracted as occurs in the case of barley.[8]

Proteins, peptone, ethylenediamine, histidine and compounds containing the group —N·C(X)·N— (where X is not oxygen) effect maximum liberation of the insoluble amylases. Partial liberation is obtained with salts, surface-active agents, and basic substances such as the monoamines, basic amino-acids and sodium hydroxide. A comparison of the extraction of the insoluble amylase of barley with those of kaffircorn malts reveals many contrasts. Peptone, an excellent extractant of the amylases of birdproof kaffircorn malt, and surface-active agents are little better than water in extracting insoluble barley amylase. Cysteine is a very effective extractant of the insoluble amylases of barley and barley malts but a very poor one for those of the kaffircorn malts. Both birdproof kaffircorn malt and barley contain moderate amounts of salt-soluble amylase, but, for the former, maximum liberation of amylase is only attained by the use of concentrations two to five times as great as are necessary for the latter.

The release of the insoluble amylases of birdproof kaffircorn malt is not a proteolytic process, inactive papain being as efficient as an active preparation. Furthermore, the release cannot be attributed to the severance of disulphide linkages, since cysteine is practically without effect on the insoluble amylases. The present work suggests that the insoluble amylases of kaffircorn malts may be adsorbed on the surface of a globulin or a nucleoprotein in such a way as to leave their active centres free to dextrinise and saccharify starch as do the soluble enzymes. The amylases are not in themselves insoluble but are held to the insoluble adsorbing substance with such firmness that aqueous extraction cannot effect their release.

There appear to be three ways in which the adsorbed enzymes may be brought into solution :

(i) by dissolving the adsorbing substance, thus leaving them free, e.g., by the use of salt solutions. This does not, however, give complete liberation of amylase. It is possible that a certain proportion of amylase is adsorbed on a protein which is not brought into solution by salts.

(ii) desorption by the use of a surface-active agent which will spread over the adsorbing surface to displace the enzyme. The yield of amylase here is slightly below the maximum.

(iii) The basic substances and those containing the —N·C(X)·N— group are postulated as releasing amylase by specific desorption at critical sites on the adsorbing surface, the —N·C(X)·N— group apparently possessing some specific configurational property which makes it particularly effective. Surface-active agents by contrast possess a more general and slightly less effective action.

The effectiveness of the proteins and peptone is probably due to both surface-active properties and the presence of basic amino-acids, particularly histidine.

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EFFECTS OF NITROGEN AND POTASSIUM FERTILISERS ON THE MINERAL STATUS OF PERENNIAL RYEGRASS (*LOLIUM PERENNE*). I.—Mineral Content

By H. RAHMAN, P. McDONALD and K. SIMPSON

Results are given for the effects of ammonium nitrate applied at two levels and potassium sulphate at one level on the nitrogen, calcium, magnesium, sodium, potassium, phosphorus, sulphur and chlorine content of perennial ryegrass grown pure and in the presence of clover. Three cuts were taken over two seasons and statistical analyses of the results are presented for each cut.

Application of nitrogen fertilisers increased the N content of the ryegrass (chiefly the non-protein nitrogen). Sodium contents were increased at all cuts and potassium in the second and third cuts, after application of nitrogen. Chlorine content was decreased and phosphorus increased (at second and third cuts). Variable magnesium contents were obtained, possibly due to rainfall conditions.

Introduction

A number of workers, including Bartlett *et al.*[1] and Brouwer,[2] have commented on the possible adverse effect of heavy dressings of nitrogenous and potassic fertilisers on grassland. These workers have stressed the probability of increasing the incidence of hypomagnesaemic tetany in ruminant animals grazing pastures overdosed with these fertilisers. Although a number of workers including Stewart & Holmes,[3] Bosch[4] and Thomas & Thompson[5] have provided valuable data on the effects of different manurial treatments on the mineral composition of grasses, there is still a considerable lack of information on the effects of fertiliser treatment with specific reference to magnesium.

It is clear that the mineral composition of a sward is dependent on many factors other than the mineral status of the soil and such factors as botanical make-up, stage of growth, and season cannot be overlooked. In this investigation the effect on grass of two levels of nitrogenous fertiliser and one of potassium has been studied in herbage sampled at three different times of the year from replicated plots. Attention has been given to a single species, namely perennial ryegrass, although the effect of the presence of clover on the ryegrass has also been studied.

Experimental

The experimental site was at Bush Estate, Midlothian. The soil of the experimental area is a sandy clay loam, derived from water-worked till with drainage impeded at 2 ft. to 2 ft. 6 in. At the time of the experiment the average soil pH was 6·0. The available phosphorus and potassium contents were low.

The design was a $3 \times 2 \times 2$ factorial with three replications, three rates of nitrogen (N_0, N_1 and N_2) being applied with and without potassium (K_0 and K). The species of grass used was S.100 perennial ryegrass. One half of the plots was sown pure (C_0) and the other half with S.24 white clover (C). The treatments were randomised in six sub-blocks so that the interaction of KC and the triple interaction NKC were partially confounded with blocks. These interactions were almost always non-significant and for the purpose of the statistics presented below the degrees of freedom allocated to them have been included with those for error.

All nitrogen treatments were applied in the form of ammonium nitrate and the potassium was applied as potassium sulphate. Table I shows the time and rate of application of fertilisers.

Table I

Fertiliser treatments in lb. of N and K/acre

Treatment	20/4/56	20/7/56	23/12/56	7/3/57
K (lb. K/acre)	188	—	188	—
N_1 (lb. N/acre)	84	84	—	84
N_2 (lb. N/acre)	168	168	—	168

The grass and clover were sown on 15/5/56 at the rates of 30 lb. and 2 lb. per acre, respectively. Two cuts were taken during 1956 on 18/7/56 and 20/8/56 and one during the following year on 23/5/57. In these experiments only grass was analysed and the samples were taken for analysis after separation from clover and weeds. Weed competition was severe in the early weeks and it was necessary to hand-weed all plots before the first cut was taken. Soil contamination was removed by rapid washing with water and the samples were placed on wire netting to drain prior to drying in an electric oven for 24 h. at 100°.

Methods of analysis

Calcium and magnesium were determined by the methods of analysis for stock feeds laid down by the A.O.A.C. ;[6] phosphorus by the method of Richards & Godden ;[7] chlorine by Caldwell & Moyer's modification[8] of Husband & Godden's method ; sulphur by the procedure of Marston[9] and sodium and potassium by the flame photometer method of Peach & Tracey.[10]

Results

Nitrogen content

The effect of treatments on the percentage of total and non-protein nitrogen is shown in Table II. In all the tables, the main effects of nitrogen, potassium and clover treatments are shown in the form of two-way tables giving also the interactions of nitrogen with both potassium and clover. L.S.D. (least significant differences) ($P = 0.05$) are given below the main table. If the differences were non-significant, no L.S.D. is given. It should be noted that the nitrogen treatments greatly affected the amount of clover in the sward, the N_0 plots having a high proportion of clover, N_1 a moderate amount and N_2 plots very little. NC interactions may, therefore, often be significant on account of the lack of clover on high-nitrogen-treated plots.

Table II

Effect of treatments on the percentage of total and non-protein nitrogen in the dry matter of ryegrass

Total nitrogen in dry matter, %

Treatment	1st cut				2nd cut				3rd cut			
	N_0	N_1	N_2		N_0	N_1	N_2		N_0	N_1	N_2	
K_0	1·65	1·68	2·20	1·84	2·53	2·93	3·27	2·91	1·80	2·48	3·08	2·46
K	1·77	1·85	2·08	1·90	2·27	2·80	3·08	2·71	1·75	2·37	2·98	2·37
C_0	1·48	1·75	2·27	1·83	2·02	2·83	3·17	2·67	1·70	2·50	2·97	2·39
C	1·93	1·78	2·02	1·91	2·78	2·92	3·19	2·96	1·85	2·35	3·10	2·43
	1·71	1·77	2·14		2·40	2·87	3·18		1·78	2·43	3·03	

L.S.D.

1st cut	2nd cut	3rd cut
Main N = 0·086	Main N = 0·121	Main N = 0·103
Main C = 0·071	Main K, C = 0·099	Main K = 0·084
Interaction NK, NC = 0·123	Interaction NC = 0·172	Interaction NC = 0·147

Non-protein nitrogen in dry matter, %

Treatment	1st cut				2nd cut				3rd cut			
	N_0	N_1	N_2		N_0	N_1	N_2		N_0	N_1	N_2	
K_0	0·29	0·38	0·56	0·41	1·00	1·54	1·58	1·38	0·53	0·72	0·92	0·72
K	0·25	0·47	0·55	0·42	0·72	1·42	1·32	1·15	0·49	0·65	0·78	0·64
C_0	0·20	0·35	0·55	0·36	0·50	1·46	1·45	1·14	0·49	0·75	0·85	0·70
C	0·34	0·51	0·56	0·47	1·22	1·50	1·47	1·39	0·53	0·61	0·85	0·67
	0·27	0·43	0·55		0·86	1·48	1·46		0·51	0·68	0·85	

L.S.D.

1st cut	2nd cut	3rd cut
Main N = 0·107	Main N = 0·112	Main N = 0·090
Main C = 0·088	Main K, C = 0·091	Main K = 0·074
	Interaction NC = 0·158	

The expected increase in the total nitrogen with increased nitrogen applications was found in all three cuts, although only the higher dressing gave a significant increase at the first cut. In both 1956 cuts, practically the whole of the increase in total nitrogen was accounted for by the highly significant increases in non-protein nitrogen brought about by application of nitrogen. In 1957, however, only about one-third of the extra uptake was in the form of non-protein nitrogen and considerable increases in true protein resulted.

Except at the first cut where it had no overall effect, the potassium treatment depressed both total and non-protein nitrogen. The reduction in non-protein nitrogen more than accounted for that in total nitrogen at both the second and third cuts. In the first cut there was an interaction between nitrogen and potassium treatments on the total nitrogen percentage, potassium treatments stimulating nitrogen uptake at the N_0 and N_1 levels and depressing it at the N_2 level.

The presence of clover along with the ryegrass increased the percentage of total nitrogen at all cuts (3rd cut not significant). This increase was also associated with one in non-protein nitrogen. The nitrogen–clover interaction was significant at all three cuts, the increase in nitrogen content of the ryegrass being associated with the presence of clover, while on the nitrogen-treated plots where clover was lacking it was non-significant. At the second cut, the increased nitrogen content of the grass, associated with the presence of clover, could be accounted for by the increase in non-protein nitrogen, but at the other two cuts the non-protein nitrogen increased by only a small fraction of the total nitrogen increase.

Table III shows the effect of treatments on the percentage of the cations magnesium, calcium, sodium and potassium in the dry matter of ryegrass at all three cuts.

Magnesium content

The effect of nitrogen applications on the magnesium content of the ryegrass altered as the sward became established. In the first cut, where weed competition was severe, the overall effect was a reduction in the percentage of magnesium particularly at the N_1 level. This picture was complicated by NK and NC interactions. The increase in magnesium content of ryegrass by nitrogen application occurred only in the presence of potassium, a depression being recorded when nitrogen only was used. Also, nitrogen applied to the pure ryegrass, and the presence of clover, depressed the magnesium content.

At the second cut, nitrogen treatments had little effect on the magnesium content but a small significant increase was brought about by the introduction of clover. At the third cut, however, on the well-established sward in 1957, increasing nitrogen treatments had a striking effect in increasing the magnesium content. For the N_1 and N_2 treatments the increase over control was about 16 and 33% and was independent of the presence of potassium fertilisers or clover.

The effect of potassium fertilisers in reducing the magnesium content of grass was found only at the first cut and then only in the absence of or at the first level of application of nitrogen.

Calcium content

Nitrogen treatments decreased the percentage of calcium in the ryegrass at the first cut. This effect was also observed at the second cut but only in the clover–ryegrass plots. There was a strong nitrogen–clover interaction at this cut and on pure ryegrass plots the nitrogen treatments strikingly increased the calcium content of the herbage. At the third cut (May 1957) nitrogen treatments increased the calcium content irrespective of the presence of clover.

Potassium treatments steadily and considerably depressed the percentage of calcium in the ryegrass. The overall effects of clover were not significant at any cut.

Sodium content

The effect of clover on the sodium content of the ryegrass was not significant at any stage, and there was no interaction of nitrogen with clover. Nitrogen and potassium treatments, however, both had very marked effects and interacted strongly.

The potassium treatments progressively depressed the sodium content from the first to the third cut, the depression being 13, 38 and 60% for the three cuts. It did not occur at any cut unless nitrogen had also been applied but was most marked at either the first or second level of nitrogen application. The sodium content of the N_2K_0 ryegrass at the third cut was in fact more than three times that from the N_2K_1 plots.

Nitrogen applications, on the other hand, strongly increased the percentage of sodium in the ryegrass at all three cuts. Again, this was not an independent effect, being stronger at all cuts in the absence of potassium treatment.

Table III

Effect of treatments on the percentage of Mg, Ca, Na and K in the dry matter of ryegrass

MAGNESIUM

Treatment	1st cut N_0	N_1	N_2		2nd cut N_0	N_1	N_2		3rd cut N_0	N_1	N_2	
K_0	0·208	0·175	0·178	0·187	0·217	0·215	0·225	0·219	0·122	0·147	0·167	0·145
K	0·153	0·163	0·170	0·162	0·208	0·217	0·212	0·212	0·122	0·143	0·162	0·142
C_0	0·192	0·165	0·175	0·177	0·200	0·200	0·217	0·206	0·123	0·148	0·162	0·144
C	0·170	0·173	0·173	0·172	0·225	0·232	0·220	0·226	0·120	0·142	0·167	0·143
	0·181	0·169	0·174		0·213	0·216	0·218		0·122	0·145	0·164	

L.S.D. Main N = 0·0085; Main K = 0·0085; Interaction NK, NC = 0·0120. Main C = 0·0075; Interaction NC = 0·013. Main N = 0·0078

CALCIUM

Treatment	1st cut N_0	N_1	N_2		2nd cut N_0	N_1	N_2		3rd cut N_0	N_1	N_2	
K_0	0·553	0·518	0·497	0·523	0·530	0·538	0·527	0·532	0·447	0·498	0·527	0·491
K	0·483	0·465	0·452	0·467	0·475	0·472	0·490	0·479	0·408	0·397	0·430	0·412
C_0	0·523	0·493	0·467	0·494	0·463	0·500	0·538	0·501	0·420	0·447	0·477	0·448
C	0·513	0·490	0·482	0·495	0·542	0·510	0·478	0·510	0·435	0·448	0·480	0·454
	0·518	0·492	0·474		0·503	0·505	0·508		0·428	0·448	0·478	

L.S.D. Main N = 0·023; Main K = 0·019. Main K = 0·152; Interaction NC = 0·026. Main N = 0·017; Main K = 0·014; Interaction NK = 0·024

SODIUM

Treatment	1st cut N_0	N_1	N_2		2nd cut N_0	N_1	N_2		3rd cut N_0	N_1	N_2	
K_0	0·083	0·128	0·218	0·143	0·060	0·135	0·207	0·134	0·052	0·130	0·227	0·139
K	0·105	0·105	0·168	0·126	0·050	0·063	0·138	0·084	0·053	0·052	0·065	0·057
C_0	0·080	0·128	0·167	0·131	0·052	0·102	0·170	0·108	0·053	0·110	0·130	0·101
C	0·108	0·105	0·203	0·139	0·058	0·097	0·175	0·110	0·052	0·082	0·152	0·095
	0·094	0·117	0·193		0·055	0·099	0·173		0·053	0·096	0·146	

L.S.D. Main N = 0·026; Interaction NK = 0·036. Main N = 0·012; Main K = 0·010; Interaction NK = 0·017. Main N = 0·016; Main K = 0·013; Interaction NK = 0·023

POTASSIUM

Treatment	1st cut N_0	N_1	N_2		2nd cut N_0	N_1	N_2		3rd cut N_0	N_1	N_2	
K_0	3·36	2·84	3·12	3·11	3·78	4·60	4·30	4·23	2·40	2·57	2·75	2·57
K	3·23	3·11	3·63	3·32	3·67	4·38	5·15	4·40	2·42	3·00	3·55	2·99
C_0	3·01	2·85	3·50	3·12	3·43	4·40	4·70	4·18	2·35	2·70	3·90	2·68
C	3·57	3·10	3·25	3·31	4·02	4·58	4·75	4·45	2·47	2·87	3·30	2·88
	3·29	2·98	3·37		3·73	4·49	4·73		2·41	2·78	3·15	

L.S.D. Main N = 0·23; Main K = 0·19; Interaction NK, NC = 0·33. Main N = 0·27; Main C = 0·22; Interaction NC = 0·38. Main N = 0·11; Main K, C = 0·12; Interaction NK = 0·21

Potassium content

At the two later cuts applications of nitrogen at both levels increased the content of potassium. The NK interaction was marked at all three cuts, the higher nitrogen treatment giving a better effect in all cases in the presence of potassium.

The application of potassium did not increase the potassium content of the ryegrass as much as was expected.

As stated above the nitrogen and potassium effects were interdependent and at all three cuts the potassium treatment increased the K content of the ryegrass most efficiently in the presence of the higher level of nitrogen.

The presence of clover had also a stimulating effect on the potassium content. This overall effect just failed to reach significance at the first cut, where it was strongest in the absence of nitrogen, but was significant at both the second and third cuts.

Table IV shows the effect of treatments on the percentage of phosphorus, sulphur and chlorine in the dry matter of ryegrass.

Table IV

Effect of treatments on the percentage of P, S and Cl in the dry matter of ryegrass

PHOSPHORUS

Treatment	1st cut N_0	1st cut N_1	1st cut N_2		2nd cut N_0	2nd cut N_1	2nd cut N_2		3rd cut N_0	3rd cut N_1	3rd cut N	
K_0	0·348	0·332	0·345	0·342	0·467	0·482	0·498	0·482	0·342	0·378	0·413	0·378
K	0·392	0·347	0·353	0·365	0·398	0·440	0·495	0·444	0·318	0·378	0·398	0·365
C_0	0·407	0·338	0·338	0·361	0·412	0·430	0·483	0·442	0·330	0·373	0·402	0·368
C	0·333	0·340	0·360	0·344	0·453	0·492	0·510	0·485	0·330	0·383	0·410	0·374
	0·370	0·339	0·349		0·433	0·461	0·497		0·330	0·378	0·406	

L.S.D. Main K, C = 0·016; Interaction NK, NC = 0·027 | Main N = 0·024; Main K, C = 0·019; Interaction NK = 0·033 | Main N = 0·019

SULPHUR

Treatment	1st cut N_0	1st cut N_1	1st cut N_2		2nd cut N_0	2nd cut N_1	2nd cut N_2		3rd cut N_0	3rd cut N_1	3rd cut N_2	
K_0	0·390	0·275	0·355	0·340	0·477	0·423	0·390	0·430	0·273	0·307	0·333	0·304
K	0·293	0·312	0·322	0·309	0·425	0·393	0·423	0·414	0·307	0·313	0·337	0·319
C_0	0·355	0·312	0·355	0·341	0·465	0·387	0·407	0·419	0·308	0·310	0·328	0·316
C	0·328	0·275	0·322	0·308	0·437	0·430	0·407	0·424	0·272	0·310	0·342	0·308
	0·342	0·293	0·338		0·451	0·408	0·407		0·290	0·310	0·337	

L.S.D. Main N = 0·029; Main K, C = 0·024; Interaction NK = 0·041 | Main N = 0·024; Interaction NK, NC = 0·034 | Main N = 0·013; Main K = 0·011; Interaction NC = 0·019

CHLORINE

Treatment	1st cut N_0	1st cut N_1	1st cut N_2		2nd cut N_0	2nd cut N_1	2nd cut N_2		3rd cut N_0	3rd cut N_1	3rd cut N_2	
K_0	0·468	0·407	0·373	0·404	0·468	0·428	0·383	0·427	0·563	0·498	0·505	0·522
K	0·588	0·393	0·508	0·508	0·597	0·498	0·377	0·491	0·632	0·568	0·502	0·567
C_0	0·590	0·407	0·417	0·471	0·518	0·460	0·337	0·438	0·618	0·522	0·517	0·552
C	0·467	0·393	0·465	0·442	0·547	0·467	0·423	0·479	0·575	0·545	0·492	0·537
	0·528	0·400	0·441		0·533	0·463	0·380		0·597	0·533	0·503	

L.S.D. Main N = 0·057; Main K = 0·047; Interaction NC = 0·081 | Main N = 0·026; Main K, C = 0·022; Interaction NK, NC = 0·037 | Main N = 0·031; Main K = 0·026; Interaction NK = 0·045

Phosphorus content

At the first cut the overall effect of nitrogen treatments on the phosphorus content was not significant but, both in the presence of potassium and in the absence of clover, nitrogen treatments depressed the amount of phosphorus in the plant.

After the sward was well established, the two applications of nitrogen steadily increased the percentage of phosphorus in the dry matter. Apart from a slight NK interaction at the second cut, this effect was independent of either potassium applications or the presence of clover.

The potassium treatment increased the phosphorus content at the first sampling particularly where no nitrogen was applied. In the second cut this effect was strikingly reversed and a depression in the percentage of phosphorus was induced by the potassium treatment, the effect being most marked in the absence of nitrogen. A similar trend was noted at the third cut but the differences were not significant.

The effect of the presence of clover in the sward on the uptake of phosphorus by the ryegrass also changed as the sward became established. A very marked depression in P content, at the first cut, was induced by the presence of clover, probably because of the vigorous competition for phosphorus particularly where the clover was strongest (N_0 plots). At the second cut, however, the presence of clover strongly increased the phosphorus content of the ryegrass. The effect at the third cut, although slightly positive, was non-significant.

Sulphur content

The sulphur content of ryegrass was depressed by the lower level of nitrogen at the first sampling and by both levels at the later sampling in 1956. In both cases the effect was dependent on whether potassium sulphate had been applied or not and did not follow a regular pattern. At the third cut the sulphur content was increased by both nitrogen treatments and, as would be expected, by the potassium sulphate dressing.

Although the presence of clover had an overall effect only at the first cut, it depressed the sulphur content of ryegrass at all three cuts in the absence of nitrogen.

Chlorine content

The effect of treatments on chlorine content was rather unexpected. Nitrogen applications at all cuts produced a continuous and most marked reduction in chlorine content. At the second and third cuts the presence of potassium enhanced this depression. The overall effect of potassium treatments was to increase the chlorine content at all cuts. This effect became less marked as the sward became established, being 25%, 16% and 10% at the three cuts. As stated above the effects of potassium and nitrogen treatments were interdependent at the later cuts, the potassium salt having a very strong positive effect at the N_0 level and no effect at all at the N_2 level.

The effect of clover on chlorine uptake followed a similar pattern to that of the other ' anion elements '—depressing the percentage at the first cut, particularly where no N was applied, stimulating it at the second cut and being ineffective at the third cut.

Discussion

Bartlett *et al.*[1] have demonstrated the effect of heavy dressings of ammonium sulphate in increasing the incidence of hypomagnesaemic tetany in cows, although the condition is not an inevitable outcome of the use of this fertiliser. The importance of nitrogen metabolism in this condition has been mentioned by a number of workers including Derivaux[11] and Head & Rook.[12] In the present experiments nitrogen applications markedly increased the percentage of total nitrogen in the ryegrass at all cuts, but in the 1956 cuts the increase was accounted for almost entirely by increases in non-protein nitrogen (N.P.N.). The increase in N.P.N. associated with application of nitrogenous fertilisers may be a significant factor in the aetiology of hypomagnesaemic tetany.

The increase in total N content was always much more marked in the pure ryegrass plots. The N content of the ryegrass grown with clover on the N_0 plots was high. This effect was not shown in the N_1 and N_2 plots because of the suppression of clover by nitrogen treatments. A considerable amount of the increase in nitrogen associated with the presence of clover was in the form of N.P.N., especially in the second cut.

The effect of nitrogen applications in increasing the sodium content was highly significant at all cuts ; this was more noticeable where no potassium had been applied. In the case of the potassium content of the grass, nitrogen applications increased the amount present only at the second and third cuts.

Since the magnesium content of herbage is regarded as being of considerable importance in the incidence of hypomagnesaemia it is necessary to consider the effects of treatments on this element in detail. The effect of high dressings of potassium fertilisers on the magnesium content of herbage has been widely investigated. Bartlett *et al.*[1] found that the application of potassium sulphate to grass tended to depress the magnesium content, although the effect on the grass was less marked when clover was present. A similar effect was observed by Stewart & Holmes[3] and by Brouwer[2] who found that the calcium content was also depressed.

Such a depression of magnesium by potassium fertilisers occurred only in the first cut in the present experiments and although it was highly significant in the N_0 plots, the significance was absent on the N_2 plots. Apart from this exception these results do not confirm those quoted above ; it is difficult to explain this.

The mean percentage values for magnesium at the three cuts were 0·17, 0·22 and 0·14. These values vary considerably and suggest that weather conditions are having some effect. The rainfall at the site for May, June, July and August, 1956, was 1·21, 2·56, 5·12 and 8·04 in. respectively. More than half of the July rainfall fell in the last three days of the month and would not affect the first cut (18/7/56). In 1957 the monthly rainfall for March, April and May was 2·24, 1·17 and 1·68 in. It is obvious from this that the two low magnesium values were associated with low rainfall conditions. The higher value was obtained for grass grown during a very wet period.

The other obvious weather factor is temperature, but it is difficult to assess the effect of this except under carefully controlled conditions since Dijkshoorn & t'Hart[13a] have found that changes of temperature over a short period of time can affect the cationic composition of ryegrass.

The depression of calcium by potassium mentioned by Brouwer[2] was very marked at all cuts. This is not surprising considering the antagonism between these two elements in the soil.

The effect of nitrogen treatments in depressing the chlorine content was most striking. Dijkshoorn[13b] has commented that nitrates will depress the absorption of chlorine, sulphur and phosphorus. In the above experiments there is no consistent depression of sulphur and at the second and third cuts the phosphorus content is significantly increased. This finding is in agreement with results obtained by Simpson[14] for other crops. Reference has been made to the possible effect of rainfall variations on the magnesium content of grass. A similar relationship exists between phosphorus content and rainfall, the phosphorus figures for the second cut being much higher than for the other two. This agrees with work carried out by Simpson[15] on grass and other crops.

Conclusions

(1) Applications of ammonium nitrate increased the percentage of nitrogen in ryegrass at all cuts but this increase was mainly accounted for by non-protein nitrogen.

(2) Ryegrass grown with clover in the absence of nitrogenous fertilisers contained more nitrogen than when grown as a pure sward.

(3) The ammonium nitrate increased the sodium content at all cuts and generally increased phosphorus and potassium, but magnesium only at the third cut. This fertiliser decreased the percentage of chlorine in the ryegrass.

(4) Applications of potassium sulphate generally increased the potassium and chlorine contents and decreased calcium and sodium at all cuts. This fertiliser also generally decreased non-protein nitrogen, but only decreased magnesium at the first cut. The presence of clover generally increased the potassium and decreased the sulphur content of the ryegrass.

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JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE

ABSTRACTS

JULY, 1960

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

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JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE

ABSTRACTS

JULY, 1960

I.—AGRICULTURE AND HORTICULTURE

General: Soils and Fertilisers

Nature and properties of major soils of the Lajas valley, Puerto Rico. M. A. Lugo-López, R. Pérez-Escolar, G. Acevedo and J. Juárez, jun. (*Puerto Rico agric. Exp. Sta.*, 1959, Bull. 149, 60 pp.).—Morphological, physical and chemical data are presented for 23 soil profiles representing six soil series. The results are discussed, particularly in relation to the feasibility of irrigation for the area. A. H. CORNFIELD.

Fertility characteristics of Oklahoma soil associations. R. M. Smith, F. Gray and H. M. Galloway (*Okla. agric. Exp. Sta.*, 1959, Bull. 528, 27 pp.).—Values for pH, exchangeable K, acid-sol. P and org. C are summarised from over 11,000 soil tests. The results are analysed and interpreted to indicate nutrient status trends for different areas and are discussed in relation to climate, parent material and vegetation. A. H. CORNFIELD.

Aluminium in interlayers of vermiculite. C. I. Rich (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 26—32).—Al sorbed from N-$AlCl_3$ in the interlayer spaces of vermiculite (0·2—2·0μ) was not entirely removed by extensive washing with neutral salt solutions. Al fixation was increased by heating the Al-clay with $AlCl_3$ solution or with water. Treatment with $AlCl_3$–NaOH reduced the exchange capacity to very low values. Most of the interlayer Al was removed by titration with NaOH in N-NaCl to pH 10. The presence of interlayer Al reduced NH_4^+ fixation, increased its stability to heat treatment and decreased its ability to expand when treated with glycol. A. H. CORNFIELD.

Cholesterol as a standard in the *X*-ray diffraction of clay minerals. J. A. Kittrick (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 17—20).—Cholesterol was a satisfactory standard for aligning *X*-ray diffractometers for work with the relatively long spacings which are the chief diagnostic features of clay minerals. Improved results with clay minerals were obtained using this method. A. H. CORNFIELD.

Weathering of fly-ash. L. H. Jones and A. V. Lewis (*Nature, Lond.*, 1960, **185**, 404—405).—Graphs show the rates of removal, by water and Morgan's reagent, of Al, Fe, PO_4^{3-}, Ca, B and K from fly-ash of varying age (0—25 years). A rapid initial decrease in concn. during the first 5 years is followed by a steady release of the elements. There is equivalence in the decreases of available Fe and PO_4^{3-} and those of available Ca and Al; interaction between Ca and Al is probable, but the rapid leaching of sol. Ca (laboratory analyses) is unlikely under field conditions. Concn. of B and Al are initially injurious to plant growth, but rapidly fall to levels for a normal soil. The range of possible crops on freely-drained fly-ash tips is limited during the first few years, but becomes wider as the ash weathers. W. J. BAKER.

Models for reactions between organic and mineral substances in the soil. R. Hess, R. Bach and H. Denel (*Experientia*, 1960, **16**, 38—40).—Models are proposed and investigated, for the reaction between org. and mineral substances in soil, which cause speedier weathering, percolation shift and crumbling of the soil. Low-mol. siloxanes, silica gel, Al_2O_3, Permutit and perlite were dissolved by ammoniacal catechol solution, and silica gel by other *o*-diphenols. Cryst. low-mol. silicon-org. ester or complex compounds were isolated from the reaction products of silica gel with catechol and 2,3-dihydroxynaphthalene. (35 references.) M. LAPIDOT.

Water-retention properties of Oklahoma soils. H. V. Eck and B. A. Stewart (*Okla. agric. Exp. Sta.*, 1959, Bull. 526, 12 pp.).—Bulk density, moisture equiv., moisture retained at 15-atm. tension, and available water capacity data for 17 soils, mainly silt loams and silty clay loams, are presented. A. H. CORNFIELD.

Water and ion movement in thin films as influenced by the electrostatic charge and diffuse layer of cations associated with clay mineral surfaces. W. D. Kemper (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 10—16).—Calculations based on the Gouy diffuse layer theory and fluid mechanics indicate a large degree of retardation of fluid and electrolyte movement in thin films of solution as a result of the presence of the adsorbed ion and electrostatic charge associated with adjacent mineral surfaces. Experimental data were in semi-quant. agreement with calculated results for Na-saturated systems. A. H. CORNFIELD.

Field infiltration studies with green manures and crop residues on irrigated soils. W. W. Williams and L. D. Doneen (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 58—61).—Gramineous summer and winter green manures improved infiltration rates in irrigation furrows in comparison with fallow soils only in medium- and fine-textured soils. Maize residues were more effective than maize roots alone or cotton residues in improving infiltration. Summer and winter annual leguminous green manures had no effect on infiltration. A. H. CORNFIELD.

Energy balance approach to evapotranspiration from crops. C. B. Tanner (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 1—9).—Micro-meteorological methods are able to estimate evapotranspiration for periods of 1 hr. or less. Evapotranspiration was estimated with good accuracy from the energy disposition at the earth's surface. Some implications of the energy balance concept with respect to consumptive-use experiments are discussed. A. H. CORNFIELD.

Evapotranspiration problems in Iraq. C. Stork (*Neth. J. agric. Sci.*, 1959, **7**, 269—282).—Actual net irrigation requirements of inundated, irrigated and drained plots were compared with empirical and theoretical methods based upon climatic conditions. A. H. CORNFIELD.

Principles and practices in the irrigation of Texas soils. M. E. Bloodworth (*Texas agric. Exp. Sta.*, 1959, Bull. 937, 55 pp.).—A comprehensive account. A. H. CORNFIELD.

Soil-conservation aspects of managing cover crops under intensive cropping systems with special reference to soil physical properties and splash erosion control. Dip Narayan Ram (*Dissert. Abstr.*, 1959, **20**, 1928).—With maize crops receiving moderate and heavy fertilisation respectively, the contents of org. matter in the soil were maintained for 7 years at 2·63 and 2·97%, with no source of org. matter other than crop residues; at 3·10 and 3·26% with cover crops, and somewhat higher with wood chips as well as a cover crop. Cover crops increased the % of water-stable aggregates, decreased the bulk density and increased the level of N in the soil, and decreased splash erosion by rain. Compaction by tractor was greater with cover crops, and spread over a larger area. The average yield of maize was less with cover crops because of poorer drainage. M. D. ANDERSON.

Fibre-glass filters for reducing silting in tile drains. V. Overholt (*Agric. Engng*, 1959, **40**, 604—607).—Tests on a fine sandy soil (in which tile drains normally silted up rapidly) with a screen of reinforced glass fibre placed around the tiles (75% of the circumference being covered) resulted in 3·5 times less silt accumulation and 1·7 times greater water discharge. A. H. CORNFIELD.

Physical and chemical changes in soil brought about by saturation with natural gas. R. S. Adams, jun. and R. Ellis, jun. (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 41—44).—Characteristics of four gas-saturated field sites were compared with those for adjacent normal soils. Gas-impregnated soils contained considerably more total C and exchangeable Mn^{2+} and Fe^{3+}, appreciably more exchangeable Fe^{2+} and generally had higher pH and available P than had the normal soils. The gas-impregnated soils also had higher water retention and total porosity and lower bulk density than had normal soils. The disturbed Fe–Mn relationships may be the major factor accounting for poor plant growth on gas-impregnated soils. A. H. CORNFIELD.

Influence of neutron meter access tube on soil temperature. R. J. Hanks and S. A. Bowers (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 62—63).—Access tubes buried in the soil for determining soil moisture content by the neutron scattering technique influenced soil temp. within 10 cm. radius of the tubes by giving lower max. and higher min. temp. than did the surrounding soil. A. H. CORNFIELD.

Determining water stability of soil aggregates. J. Miczyński (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E**, 151—172).—Current methods are reviewed and two simple, quick methods of determining water stability of soil aggregates presented. In the first method sifted aggregates, counted and placed symmetrically on filter papers, are treated with a mixture of water and acetone. The no. of whole, partly and completely broken up aggregates are noted and from these the water stability index and soil-puddling index can be calculated. The second method consists of watching the disintegration of air-dried crumbs in distilled water and in 25% ethyl alcohol during 3 min., the analysis being carried out in a simple

apparatus based on a funnel and wire sieves. Water stability is calculated from $T = (A + B)$ where A = % crumbs not broken up in water and B = % crumbs not broken up in alcohol.
E. G. BRICKELL.

Comparison of physical properties of soil after five years lucerne in mixtures with grasses and in pure cultures, and their value as preceding crops. F. Pawlowski (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E**, 197—216).—The kind of mixture had no influence on total no. of water-stable soil aggregates or vol. wt. of soil after harvest but did influence sp. gr. and hygroscopicity of the soil. The greatest amount of crop residue (roots and stubble) was accumulated by lucerne mixed with meadow fescue, and the smallest by lucerne in pure culture. The best preceding crop for wheat appeared to be lucerne with timothy grass. (25 references.)
E. G. BRICKELL.

Aluminium fixation in a synthetic cation-exchanger. P. H. Hsu and C. I. Rich (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 21—25).—Changes in cation-exchange capacity, exchangeable Al and exchangeable Na were followed during titration with NaOH of Al-saturated cation-exchange resin in absence and in presence of $AlCl_3$ and in the presence of varying mol. ratios of $AlCl_3$ and NaOH. The only exchangeable Al ion present was Al^{3+} and, in the acid range, non-exchangeable Al was fixed in the ratio one Al ion to one positive charge of the resin. A gibbsite-like ring structure having 6 Al ions, 12 OH ions and 12 water mol. is proposed as the principal fixed hydroxy-Al polymer.
A. H. CORNFIELD.

Autotrophic oxidation of ammonium and hydroxylamine. M. S. Engel and M. Alexander (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 48—50).—Oxidation of NH_2OH by *Nitrosomonas europaea* in culture tests increased with pH (6·0—9·0) and was affected by the relative concn. of substrate and bacterial cells. Nitrification of NH_4^+-N was retarded by increasing pH (8·0—9·5) particularly at the higher concn. of NH_4^+. NH_4^+ oxidation was not stimulated by addition of Fe, PO_4^{3-}, HCO^-_3, Mg, Cu or Zn, but was inhibited by 0·0001M-KCN and -allylthiourea.
A. H. CORNFIELD.

Fate of ammonia applied to soils. J. H. Baker (*Dissert. Abstr.*, 1959, **20**, 1501).—At optimal water content, immediate losses of anhyd. NH_3 applied to soils were negligible. When soils were saturated with NH_3, and then aerated for 1 week, retention of NH_3 was greatest in acid soils containing large amounts of org. matter. About 25% of the NH_3 was fixed in a form not extractable by 10% NaCl. The NH_3-retention capacity of a soil depends primarily on the amount of exchangeable hydronium and Al ions; the usual NH_4 acetate method for determining the exchange capacity of soils gave low results in many cases. NH_3 was added to silt loam as anhyd. NH_3, NH_4NO_3, $(NH_4)_2SO_4$ or $NH_4H_2PO_4$, at the rate of 500 lb. of N per acre, and water equivalent to 2·6 in. of rain was then applied. Loss of NH_4^+-N by leaching was less than 3% from soil of neutral pH. With soil of acid pH, as much as 25% of NH_4^+-N was lost from the nitrate and sulphate, but less than 3% from anhyd. NH_3 and $NH_4H_2PO_4$.
M. D. ANDERSON.

Fertility investigations on the black-earth wheat lands of the Darling Downs, Queensland. III. Mineral nitrogen in the soil: relation to the wheat crop. S. A. Waring and L. J. H. Teakle (*Aust. J. agric. Res.*, 1960, **11**, 27—41).—Data for the NO_3^-- and NH_3-N in these soils to depths of 4 ft. during fallow and cropping periods are recorded. Uptake of mineral N occurred largely from the 0—2 ft. depth of soil. In general N is not to be regarded as an important factor limiting wheat growth in this district.
A. G. POLLARD.

Effect of nitrogen volatilisation on soil acidity changes due to applied nitrogen. A. E. Hiltbold and F. Adams (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 45—47).—Soils were incubated with various sources of inorg. N + glucose under non-waterlogged conditions for 7 weeks and analysed for changes in pH, inorg. and org. N. Loss of N by volatilisation (as NH_3, oxides of N or N_2), extent of nitrification and changes in pH during incubation varied considerably. Comparison of calculated acidity (that arising from nitrification + acidity of the residual anion) with actual acidity developed during incubation showed a deficiency of actual acidity with most of the samples. This deficiency in acidity was highly correlated with the amount of N lost by volatilisation and was independent of source of N supplied.
A. H. CORNFIELD.

Dissimilar nitrifying capacities of soils in relation to losses of applied nitrogen. F. E. Clark, W. E. Beard and D. H. Smith (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 50—54).—Forty-one arable soils were incubated after addition of urea-N (400 p.p.m.). N deficit (N not recovered as NH_3, NO_2^- or NO_3^-) ranged from 2–40% of the N applied and was usually higher for poorly buffered soils, those which were initially acid or developed acidity during incubation, and those which accumulated NO_2^- or showed incomplete nitrification of NH_4^+. Incubation of the soils with NO_2^--N (400 p.p.m.) resulted in increasing deficit of applied N with decreasing soil pH and a tendency for accumulation of NH_3. Incubation of soils with added NO_3^--N resulted in no deficit of N, indicating fully aerobic conditions during all incubations and showing that deficits occurring during incubation with urea and NO_2^- were not due to enzymic denitrification. The mechanism of the losses of N from aerobic soils is discussed; probably these arise mainly from the breakdown of HNO_2 to NO or from the reaction between HNO_2 and amino-acids or NH_3 to give N_2.
A. H. CORNFIELD.

Chemical and biological changes effected in certain Ohio soils by partial sterilisation and plant growth relationships. J. M. Lapensee (*Dissert. Abstr.*, 1959, **20**, 1918—1920).—Steaming water-saturated soils for 20 hr. under 20 lb. pressure increased the solubility of Ca, Na, Mn, Mg, S and P and their uptake by crops; the solubility of K was not affected, and uptake was lessened. After short incubation NH_4^+-N was higher in steamed than in untreated soils. Sol. Mn was also increased in soils treated with dichloropropene or chloropicrin, but not so much as in steamed soils. After incubation for 6 weeks, NH_4^+-N and NO_3^--N increased in untreated and steamed soils, but decreased in those treated with chemicals. Addition of starch or maize fodder or Ca to unsteamed soil decreased available Mn. Addition of NH_4NO_3 or casein or peptone increased water-sol. Mn. Available Mn increased with the water content of the soil to 20%, decreased at 40% level, and increased again above 40%. Oats, maize and sugar beet (but not soya-beans) grew better in steamed than in untreated soil. Mn in a culture solution was toxic to some varieties of soya-bean at 10—30 p.p.m. Increase of P lessened the toxicity of Mn.
M. D. ANDERSON.

Reaction of cyanide in soils and its effect on growth and ion uptake of maize. A. H. Hunter (*Dissert. Abstr.*, 1959, **20**, 1520).—Methods and apparatus were developed for determining cyanide in soil extracts, after separation by distillation with tartaric acid. Varied amounts of KCN were removed from solution by different soils. Equilibrium was established in 24 hr., and CN^- remaining in solution was present in both free and complexed forms. The CN^- content of soils increased during the decomposition of cyanogenetic plants. Maize plants in different soils were killed by different levels of soil CN^-. In sand culture, sublethal amounts of CN^- caused changes in the P, K, Ca, Mn, Fe and Zn contents of maize plants; CN^- added to soils had somewhat different effects, varying with type of soil.
M. D. ANDERSON.

Sulphur content of Oklahoma soils, rainfall and atmosphere. H. J. Harper (*Okla, agric. Exp. Sta.*, 1959, Bull. 536, 18 pp.).—Data are given for total S and N in 170 paired samples of virgin and adjacent cultivated soils, S, N and P in 18 profiles, and SO_4^{2-} in 11 profiles to 7 ft. Loss of total S averaged 30% and of total N 37% due to cultivation. Mean N/S ratio was 7·3 in the surface layer and either remained constant or decreased with depth of profile. Total S content of rainfall ranged from 6 to 18 lb. per acre per annum.
A. H. CORNFIELD.

Soil salinity as a disturbing factor in field experiments: its significance in germination, growth and yield. K. Kreeb (*Ber. dtsch. bot. Ges.*, 1959, **72**, 123—137).—Differences in sol. salt content markedly affect the uniformity of crops in field trials. Experimental data record the effects of salinity (0·1—1·6 to salt content) on barley, wheat, maize and clover. Germination, rate of growth and yield were all affected, osmotic phenomena being mainly concerned although direct toxic effects cannot be excluded.
A. G. POLLARD.

Organic matter decomposition and plant nutrient release from incorporations of soya-bean hay and wheat straw in a Holston sandy loam in outdoor lysimeters. W. M. Shaw and B. Robinson (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 54—57).—A sandy loam (pH 4·95) was limed to 60% and 100% base saturation, treated with soya-bean hay and wheat straw and placed in outdoor lysimeters for 3 years. Org. C and total N decreased with time, but there was a net increase, in comparison with untreated soil, in both due to org. matter treatment at the end of 3 years. Results were similar at all levels of base saturation. There was less loss of org. C and total N when the soya-bean hay was mixed with the upper soil layer than when mixed with the whole soil. Losses of elements through lysimeter leachates are also reported. These losses were closely related to amounts of nutrients added in the org. materials.
A. H. CORNFIELD.

Leaching and decomposition of litter. I. Leaf litter of *Fraxinus excelsior*. II. Needle litter of *Pinus sylvestris*. N. Nykvist (*Oikos*, 1959, **10**, 190—211, 212—224).—I. Fresh (one-day-old) litter contains about 20% of water-sol. matter. K is easily leached from the litter, with P, Ca and N (in that order) less easily lost. Decomposition results in the production of water-sol. substances, but litter from which water-sol. substances have been leached decomposes slowly. During aerobic decomposition the pH rises but falls during anaerobic decomposition.

II. Less than 1% of the dry weight of the litter is water sol. Decomposition is slightly more rapid under aerobic than under anaerobic conditions and during aerobic decomposition the pH rises rapidly. Leaching of the litter does not retard decomposition. L. G. G. WARNE.

Changes in pH of leaf litter during a field experiment. H. Sjors (*Oikos*, 1959, **10**, 225—231).—Fresh litter, collected by shaking the trees, was placed on soil for 34 days. The pH of water extracts of the litter rose during this period for 11 out of 12 species (*Acer platanoides* was the exception). L. G. G. WARNE.

Wet-combustion apparatus and procedure for organic and inorganic carbon in soil. L. E. Allison (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 36—40).—Total C is determined by digestion with H_2SO_4–H_3PO_4–$K_2Cr_2O_7$, the evolved gases being passed through traps containing 50% KI, saturated Ag_2SO_4 (to remove Cl^-), conc. H_2SO_4 (to remove water), Zn (to remove acid fumes), Anhydrone (to remove traces of moisture) and the CO_2 absorbed on "Mikohbite" or "Caroxite" (self-indicating granules) and determined gravimetrically. Inorg. C is determined by digesting the soil with 2N-H_2SO_4 containing 5% $FeSO_4$ (antioxidant). A. H. CORNFIELD.

Effects of sawdust mulches. I. Soil properties. R. K. Kirsch (*Oregon agric. Exp. Sta.*, 1959, Tech. Bull. 49, 16 pp.).—Douglas fir sawdust incorporated in the soil at a rate equiv. to a 3 in. depth of sawdust had no effect on soil pH after 4 years, whereas pH decreased with increasing rate of application of $(NH_4)_2SO_4$ (**I**). Both sawdust and **I** increased total soil N but decreased nitrifiable N. Oxidisable org. matter and available soil P were decreased by sawdust, but not by **I**. Neither treatment affected available soil K. Maize leaf-N values were decreased by sawdust and increased by **I**, whilst the reverse held for leaf-P. In another test incorporation of fir and alder sawdust with the soil favourably affected aggregation, non-capillary porosity and bulk density, whilst fir sawdust applied as a surface mulch had the opposite effect. Surface mulching facilitated weed control and harvesting of strawberries. A. H. CORNFIELD.

Effects of sawdust mulches. II. Horticultural crops. A. N. Roberts and W. M. Mellenthin (*Oregon agric. Exp. Sta.*, 1959, Tech. Bull. 50, 34 pp.).—Fir and alder sawdust mulches improved the performance of blueberries to a greater extent than did straw or oak leaf mulches. Raspberry yields were not affected by any of the treatments. Sawdust mulches increased strawberry yields under disease-free conditions, but application of sawdust and/or N increased the incidence of red-stele disease. Fertiliser requirements of strawberries were not increased by surface mulches of straw, but when sawdust (60 tons per acre) was mixed with the surface soil extra N at the rate of 6·6 lb. per ton of sawdust was required in the first year, and smaller amounts in the following years. A. H. CORNFIELD.

Autotrophic bacterium oxidising ferrous iron and elemental sulphur in acid media. N. A. Kinsel (*Dissert. Abstr.*, 1959, **20**, 1533—1534). —An autotrophic bacterium isolated from bituminous coal mine drainage was a Gram-negative, non-sporing, motile rod, oxidising Fe^{2+} and elemental S in acid media. $(NH_4)_2SO_4$ was a better source of N than KNO_3, urea or peptone. Oxidation of Fe^{2+} was optimal at pH 2·85 and 32° and was favoured by shaking, by presence of vitamins and trace metals, and by an atm. containing 10% of CO_2. The organism resembled *Thiobacillus thio-oxidans*, *T. ferro-oxidans* and *Ferrobacillus ferro-oxidans*, except in its ability to oxidise both Fe and S. M. D. ANDERSON.

Effects of water and anaerobic conditions on *Fusarium oxysporum* f. *cubense* in soil. M. Newcombe (*Trans. Brit. mycol. Soc.*, 1960, **43**, 51—59).—This fungus, the cause of a serious disease of bananas, can survive in "alien" soil for at least 8 months. High CO_2 concn. and flooding of the soil both inhibit chlamydospore production and lead to a gradual reduction in activity of the fungus in soil. L. G. G. WARNE.

Physical environment of soil animals. N. Collis-George (*Ecology*, 1959, **40**, 550—557).—The energy-moisture content relation of soils is considered, together with other aspects of soil water, as well as of soil atm., soil texture, and soil temp. as they affect soil animals. L. G. G. WARNE.

Maintenance of soil fertility in the S. Guinea zone of N. Nigeria (Tiv Country). E. B. Dennison (*Trop. Agric.*, 1959, **36**, 171—176).—Fallows, cattle-pen manure and artificial fertilisers all gave worthwhile results. Short-term green manure crops were a failure. E. G. BRICKELL.

Application of soluble fertilisers through an irrigation system. I. Effect of nutrient sources and methods of fertiliser application on vegetable crops. II. Movement of soluble salts in porous media. R. H. Ruf, jun. (*Dissert. Abstr.*, 1959, **20**, 1503).—I. The effect of liquid or granulated fertiliser bands, and of split applications of N or a complete fertiliser on yields of tomatoes, potatoes, cabbage and snap-beans were investigated. Sprayed fertiliser containing free NH_3 damaged foliage and decreased yields.

II. The movement of $NaNO_3$ in columns of soil, artificial soils and sands were studied. The actual nitrate distribution in the soil at a given depth closely approximated to that calculated from the theoretical equation given. O. M. WHITTON.

Fertilisers. Fisons Ltd. (Inventors: T. P. Dee, R. E. Jewell and R. J. Nunn) (B.P. 810,208, 10.8.55 and 4.2.56).—Fertiliser of high phosphate availability (mostly in water-sol. form) is obtained by interaction of phosphate rock, viz., calciferous phosphate rock with $\leqslant$4·8 (6) mol. of NH_4 or alkali metal H sulphate in presence of water (25—200 wt.-% on rock) and/or acid, e.g., 70% aq. H_2SO_4. Thus, a mixture of water, $KHSO_4$ and Moroccan phosphate rock (80% of $<$100 in. mesh) is maintained at 20°, during 24 hr., to give a product which in water is 84·4% sol. (or 97% after 3 weeks). F. R. BASFORD.

Fertiliser containing urea and ammonium sulphate with a high nitrogen content. Bergswerkgesellschaft Hibernia A.-G. (B.P. 810,760, 12.7.57. Ger., 3.8.56).—A fertiliser product of high N% is obtained by adding NH_3 and H_2SO_4 to aq. urea (optionally already containing NH_3) at $\geqslant$140°, such that most of the water present is removed by the heat of neutralisation. F. R. BASFORD.

Fertilisers. Lummus Co. (B.P. 808,664, 21.3.56. U.S., 29.3.55). —Particulate superphosphate of $\leqslant$1 wt.-% of free acid and $\leqslant$4 wt.-% of water, especially green superphosphate, is admixed with granular Ca, K or Na metaphosphate (in excess of that needed for complete hydrolysis with added water), then the mixture is pile cured during 5 days to 2 months (after drying at 150—300° F), to provide a granular mixed fertiliser (containing 10—90 wt.-% of superphosphate). F. R. BASFORD.

Plant Physiology, Nutrition and Biochemistry

Effects of seed environment during maturation on seedling growth. F. Stearns (*Ecology*, 1960, **41**, 221—222).—Seed of *Plantago aristata* matured at 24° produced seedlings with larger cotyledons and leaves than seed matured at 15·5°, when grown at 15·5° or 24°. L. G. G. WARNE.

Control of flowering in *Indigofera endecaphylla* by photoperiod regulation. E. J. Britten and H. M. Laude (*Trop. Agric.*, 1959, **36**, 150—155).—This species exhibits short-day photoperiodic behaviour. Flowering is enhanced by advanced physiological maturity and may be prevented by immaturity. Temp. does not appear to affect flowering. E. G. BRICKELL.

Root cation-exchange capacity in explaining rootstock influence on cation content of citrus. A. Wallace (*Univ. Cal. Los Angeles, Spec. Rep.*, 1958, No. 1, 129—135).—A mathematical consideration. The ratio of cations absorbed by the roots is influenced by the cation-exchange capacity of the roots. A. G. POLLARD.

Kinetic analysis of phosphate absorption by excised roots of millet, barley and lucerne. J. C. Noggle and M. Fried (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 33—35).—Specific reaction rate constants and apparent dissociation constants are presented for P absorption by excised roots of millet, barley and lucerne. A. H. CORNFIELD.

[A] **[Effect of] girdling and cutting the xylem on absorption and translocation of nitrogen in small citrus trees.** [B] **Influence of dark on absorption and translocation of ^{15}N-tagged nitrogen in small sweet-orange seedlings.** A. Wallace and R. T. Mueller (*Univ. Cal. Los Angeles, Spec. Rep.*, 1958, No. 1, 139—143, 144—145).—[A] In soil cultures of citrus receiving ^{15}N-labelled $Ca(NO_3)_2$ severance of the xylem prevented translocation of N above the cut. Bark girdling, stem cooling and lowering soil temp. restricted N translocation.

[B] Transferring the seedlings to darkness prevented translocation of N into leaves but only slightly affected that into stems. A. G. POLLARD.

Urea and other substances in iron and zinc foliage sprays. A. Wallace and A. A. Bedri (*Univ. Cal. Los Angeles, Spec. Rep.*, 1958, No. 1, 174—178).—The Fe content of leaves of avocado, orange and soya-bean sprayed with chelated Fe [ethylenediaminedi-(*o*-hydroxyphenylacetic acid)] was increased by addition of urea (1), glycerol (1) or a herbicidal oil (1%). Urea produced the opposite effect with other Fe chelates. The oil lowered the intake of Zn from several sources. Zn chelated with dihydroxyethyl-ethylenediaminediacetic acid was absorbed by avocado leaves as readily as was that from $ZnSO_4$ but that from the EDTA chelate was absorbed only half as readily. Within the plant translocation of Fe was as rapid as that of Zn. A. G. POLLARD.

Physiological nature of sweet potato plants. IX. Variations of chemical components in the shoots and roots during the period from planting to root tuber formation. J. Naka and K. Tamaki (*Kagawa Daigaku Nôgakubu Gakuzyutu Hôkoku* [*Tech. Bull. Fac. Agric. Kagawa Univ.*], 1959, **11,** 7—12).—A study was made of sweet potato plants (var. Nôrin No. 1) during the whole growing period. Determinations were made of reducing sugar, non-reducing sugar, starch, total carbohydrate, soluble N, protein N and total N in leaf blades, petioles and vines during growth. With the growth of the plant, the carbohydrate contents in the apical parts of the vines and in the leaf blades and petioles decreased gradually up to 20—30 days after planting, but were high at the other stages. The carbohydrate contents in the basal parts of the vines were high at 20—30 days after planting, and were low in the leaf blades and petioles by 10—30 days. The carbohydrate contents in the roots increased gradually throughout. In the apical parts of the vines, a decrease in sol. N and an increase of protein-N occurred after planting, but values in the leaf blades and petioles were somewhat low 10—20 days after planting. The N contents in the basal parts of the vines were high 30 days after planting, while those in the leaf blades, petioles and roots declined gradually throughout growth. S. KAWAMURA.

Physiological nature of sweet potato plants. X. Physiological differences between the root tubers at various stages of development. XI. Variations of chemical components in the various region of seed tubers during storage. XII. Physiological behaviour of roots in tuber formation. J. Naka and K. Tamaki (*Nippon Sakumotsu Gakkai Kiji* [*Proc. Crop Sci. Soc. Japan*], 1958, **27,** 95—96, 305—306; 1959, **28,** 124—125).—X. Determinations were made of the activities of phosphorylase, phosphatase and amylase, and the ascorbic acid and carbohydrate contents in the tubers at different growth stages of sweet potato plants. The phosphorylase and phosphatase activities were lowest in the fibrous roots (**I**), intermediate in the pencil-like roots (**II**), and highest in the tuberous roots (**III**); the activities increased in **III** with increase in the thickness of the tubers. The amylase activity was lowest in **I**, highest in **II**, and intermediate in **III**. The amylase activity decreased in **III** with the growth in thickness of the tubers. The ascorbic acid (total and reduced) contents were lowest in **I**, intermediate in **II** and highest in **III**; they increased with increase in thickness of the tuber. Oxidised ascorbic acid was lowest in **I**, highest in **II** and intermediate in **III**; decreasing with increase in tuber thickness. The total sugar and starch contents were lowest in **I**, intermediate in **II** and highest in **III**. The content of total sugar decreased and that of starch increased with the thickness of the tubers.

XI. Carbohydrate and N contents at the apex, centre and tail in the pith and cortex of the seed tuber were examined. In the early stages of storage, the reducing sugar (*R*) content rose at the apex and centre both in the pith and cortex, and fell at the tail particularly in the pith. The non-reducing sugar (*NR*) content increased in all the parts of the tuber, and the starch content fell in all parts except the pith of the tail. The *R* and *NR* contents were high only in the tail pith, but low in the other parts of the tuber. The starch content was relatively high at the apex and centre, while it decreased both in the pith and cortex of the tail. In the later stages of storage, the *R* content fell at the apex, but increased considerably at the centre and tail both in the pith and cortex. The *NR* content was high at the apex and centre, and low at the tail. The starch content showed a striking contrast to the behaviour of the *NR* content in the pith and cortex. The sol. N content was generally high in the early and later stages of storage, while the protein-N content was high in the middle stage in all the parts of tuber.

XII. Possible relationships between the carbohydrate content and phosphorylase and amylase activities in sweet potato roots (var. Okinawa No. 100) and the differentiation of root tuber were examined. By the 10th day after planting, the carbohydrate content in the tuberous roots showed no particular difference from that in the fibrous root. By the 20th day, the accumulations of *NR* and starch were observed in the slightly thickened part of tuberous roots. By the 30th day, the *R*, *NR* and starch contents were high in the distinctly thickened part of tuberous roots, while the *NR* and starch were high in the upper part of tuberous roots. The carbohydrate contents in the tuberous roots apparently increase in parallel with the progress of root tuber formation; throughout tuber formation the carbohydrate contents in the lower part of tuberous roots were similar to those in the fibrous roots. The phosphorylase and amylase activities in the tuberous and fibrous roots were different and their activities in the tuberous roots were closely related to root tuber formation. S. KAWAMURA.

Nitric-perchloric acid oxidation for sulphur in plant and animal tissues. W. M. Shaw (*J. agric. Fd Chem.*, 1959, **7,** 843—847).—The effects of different conditions of time and temp. and digestion vessels in the HNO_3–$HClO_4$ oxidation of plant and animal materials have been critically examined and a procedure is described which enables the same digest to be used for total S determination as is used for mineral constituents. S recoveries in standard materials were within 2% of theoretical values and the results were comparable in accuracy and precision with those given by the A.O.A.C. method using $Mg(NO_3)_2$. S. C. JOLLY.

Histopathological effects of ozone on plant foliage. M. C. Ledbetter, P. W. Zimmerman and A. E. Hitchcock (*Contr. Boyce Thompson Inst.*, 1959, **20,** 275—282).—Foliage injury appeared as dark stipples, light flecks, necrotic patches and general chlorosis. In descending order of susceptibility the plants were tomato, bean (Pinto), tobacco (Maryland Mammoth), spinach, potato, smartweed, stevia, tobacco (Vamorr 48), chrysanthemum (Pippin), hypericum, lucerne, groundnut, tobacco (Turkish), sweet potato, sensitive plant, grape, verbena, chrysanthemum, Jerusalem cherry, avocado, sugar-beet, strawberry, mint and geranium. Gladiolus, kalanchoe, pepper, bean (Tender green) and piggy-back plant were not visibly injured by concn. up to 1 p.p.m. (22 references.) E. G. BRICKELL.

Effects of air pollution on *Lupinus* in the Los Angeles area. D. B. Dunn (*Ecology*, 1959, **40,** 621—625).—At least two constituents of "smog," one water-sol. and the other water-insol., appear to be phytotoxic. Local subspecies of *Lupinus* in Los Angeles are more resistant to "smog" than are subspecies from other parts of California. L. G. G. WARNE.

Direct uptake of fission products on ryegrass. A. Morgan (*A.E.R.E.*, 1959, R 3181, 10 pp.).—In rye-grass, grown on vermiculite (plus nutrients) and exposed to fall-out for 30 days, the direct uptake of fission products can be expressed quant. (as % of deposited activity) as ^{144}Ce 95, ^{95}Zr 50, ^{90}Sr 39, ^{89}Sr 32 and ^{137}Cs 26. Uptake is max. for the batch having highest dry-wt. of rye-grass, whilst the ^{89}Sr : ^{90}Sr ratio is less than that in rain-water collected over the same period. Although pathological effects due to ingestion of grass by cows would be negligible, such direct uptake of all fission products should be considered in the event of an accidental release of radioactivity. (14 references.) W. J. BAKER.

Irradiation as an aid in fruit variety improvement. I. Mutations in the peach. L. F. Hough and G. M. Weaver (*J. Hered.*, 1959, **50,** 59—62).—Peach trees treated with γ-rays (^{60}Co) produced mutants in which season of ripeness, fruit firmness and texture and the free stone condition had been changed. L. G. G. WARNE.

Factors affecting uptake of radioactive caesium by lettuce, grass and lucerne. E. B. Fowler and C. W. Christenson (*J. agric. Fd Chem.*, 1959, **7,** 847—849).—Increase in the available K in the soil resulted in lower ^{137}Cs/K ratios in grass, lettuce and lucerne plants. Discrimination factors against ^{137}Cs were in the range 300—1400 depending on the type of soil and plant. This discrimination may be related partly to the ion-exchange capacity and partly to the exchangeable-K content of the soil. K fertilisers may lower the ^{137}Cs/K ratio in the plant and also mobilise Cs in some soils and render it more available to the plant. S. C. JOLLY.

Effect of an auxin and an antiauxin on ammonium nitrate toxicity to citrus roots. A. Wallace and J. Bennett (*Univ. Cal. Los Angeles, Spec. Rep.*, 1958, No. 1, 146—149).—In sand-cultured seedlings the toxicity of NH_4^+-N was not reversed by the antiauxin α-1-naphthylmethylmercaptopropionic acid (**I**) at pH 4, nor did indolylacetic acid produce any additional effect. At pH 7, **I** increased the top/root ratio in plants receiving a high level of N as NH_4^+ or NO_3^-, but with low-level N at this pH the effect was reversed. A. G. POLLARD.

Effect of nitrogen, antiauxin and aeration on avocado seedlings grown in solution culture. A. E. Richmond and A. Wallace (*Univ. Cal. Los Angeles, Spec. Rep.*, 1958, No. 1, 149—154).—The antiauxin, α-1-naphthylmethylmercaptopropionic acid, increased root growth in avocado seedlings grown in high-N solution cultures, but lowered it in low-N cultures. The top/root ratio was lowered by aeration of the nutrient and by diminution in N supply. A. G. POLLARD.

New plant regulators that exude from roots. John W. Mitchell, B. C. Smale and W. H. Preston, jun. (*J. agric. Fd Chem.*, 1959, **7,** 841—843).—The *m*-chloro- and *m*- and *p*-fluoro-derivatives of α-methoxyphenylacetic acid were effective plant-growth regulators which were exuded in detectable amounts from the roots of treated plants. Four other ring-substituted deriv. were active plant-growth regulators, but these were not exuded. Exudation by roots was governed by the no. and position of substituents in both the aromatic and aliphatic portion of the mol. of the parent acid. S. C. JOLLY.

Biochemical and physical considerations relating structure to plant-growth-regulating activity of *N*-arylcarbamates, substituted ureas and chlorophenoxyacetic acids. V. H. Freed (*Dissert. Abstr.* 1959, **20,** 1562—1563).—Substitution in the 4 position had the greatest influence on the growth-regulating activity of substituted phenoxyacetic acids and ureas, and substitution in the 3 position on the activity of *O*-alkyl *N*-arylcarbamates. I.r. absorption in the region of 12$m\mu$ correlated well with the biological activity of the phenoxyacetic acids. Adsorption on proteins is the primary step in the action of growth-regulators, with modification of the structure of the protein and its enzymic activity.
M. D. ANDERSON.

Effect of certain benzazoles on higher plants. M. J. Klingensmith (*Dissert. Abstr.*, 1959, **20,** 1555—1556).—Benzimidazole (**I**), benzthiazole (**II**), benztriazole (**III**) and other benzazoles, repressed elongation of the primary root of cucumber, **II** being about 10 times as active as **I** or **III**. Adenine did not reverse this inhibition, but supplemented it in the cases of **I** and **II**. **III** in root medium inhibited internodal elongation in several genera of plants, and stimulated axillary development. **II** induced development of adventitious roots. All benzazoles caused modifications of tomato pith unlike those seen with other growth regulators. In several genera, they decreased the permeability of the root to water. **I** increased the K, Ca and Na uptake of barley roots; uptake of Cl was inhibited. M. D. ANDERSON.

Preparation and plant growth-regulating activity of crude protein hydrolysate derivatives of (±)-2-(2,4-dichlorophenoxy)propionic acid. C. F. Krewson, J. F. Carmichael, P. S. Schaffer, B. C. Smale and J. W. Mitchell (*J. agric. Fd Chem.*, 1959, **7,** 837—841).—The prep. and plant-growth-regulating properties are reported of the products resulting from the interaction of (±)-α-2,4-dichlorophenoxypropionic acid with cheap protein hydrolysates from by-products and readily obtainable protein sources in an attempt to produce amide herbicides of low volatility. Generally, the response of bean, sunflower and barley plants to all deriv. was less than to the parent acid, while that of cucumber and maize plants was greater. The reduction of undesirable formative effects characteristic of phenoxy-parent acids may be of interest for other purposes. S. C. JOLLY.

Action of alkyl *N*-phenylcarbamates on photolytic activity of isolated chloroplasts. D. E. Moreland and K. L. Hill (*J. agric. Fd Chem.*, 1959, **7,** 832—837).—All alkyl *N*-phenylcarbamates highly active in inhibiting the photolytic activity of isolated chloroplasts of turnip tops had a free imino-H atom. This inhibitory activity was lost, or decreased, on replacing this H atom by an alkyl or aryl group. Possible rôles of various substituents of the carbamate mol. in inhibiting the photolytic action are discussed. Max. herbicidal activity occurred with the propyl and butyl esters of *m*-halogenated compounds. (29 references.)
S. C. JOLLY.

Mechanism of the action of isopropyl *N*-phenylcarbamate (IPC) in preventing the sprouting of potatoes. W. Nultsch (*Phytopath. Z.*, 1959, **37,** 75—108).—The action of IPC and the corresponding ethyl derivative (EPC) in suppressing root development in potato is primarily located in the zone of active cell division of the root tip; zones of cell elongation and of differentiation are much less affected. The extent of inhibition of sprouting is directly related to the concn. of the applied substance and to the length of the period of treatment. IPC is more active than EPC in this respect. Low concn. of IPC applied for relatively short periods may stimulate sprouting, EPC being the more effective. The effects of the carbamates were not reversed by purines, thymine or thiol-compounds.
A. G. POLLARD.

Chromatography of indol-3-ylacetic acid. H-D. Klämbt (*Ber. dtsch. bot. Ges.*, 1959, **72,** 185—187).—Some modifications of technique are indicated. A. G. POLLARD.

Effect of gibberellic acid on seeded grapes. S. Lavee (*Nature, Lond.*, 1960, **185,** 395).—Grapes sprayed ~23 days after full bloom with gibberellin (20 p.p.m.) increase considerably in wt. up to full maturity, but the added wt. decreases with increasing no. of seeds in the berry. Treated/control wt.-ratio rises to ~2·5 for seedless fruit, but is only ~1 for 3-seed berries. α-Naphthylacetic acid does not affect fruit size. W. J. BAKER.

Effect of gibberellic acid and nitrogen on winter growth of pasture. R. S. Scott (*N.Z.J. agric. Res.*, 1959, **2,** 1203—1210).—A 2-year-old ryegrass (dominant)–white clover sward showed noticeable stimulation of growth with yellowing of herbage 1 week after application of gibberellic acid (0·9 oz./acre). In 2 weeks, growth was twice that of untreated plots. Stem elongation and narrowing of leaf blade occurred in grasses; clovers showed increase in length of stem and leaf area. Fertiliser-N acted more slowly, but 6 weeks after its application grass production equalled that from gibberellic acid. Total-N in acid-treated grass increased up to 6 weeks but was lower after 14 weeks. (11 references.) K. R. BUTLIN.

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Increased development of xylem in *Solanum nigrum* due to gibberellic acid. O. Kiermayer (*Ber. dtsch. bot. Ges.*, 1959, **72,** 343—348).—The K salt of the acid was applied to *S. nigrum* seedlings in the 6-leaf stage, at the rate of 2—200 μg./plant. Xylem development at the internodes was considerably increased; the new cells produced were normal and lignified to similar extents as in untreated plants. The no. of lignified cells formed increased with the dosage and the effect was apparent, to decreasing extents, in the higher internodes. A. G. POLLARD.

Crops and Cropping

Relationship between wheat yields, available moisture and available nitrogen in dry-land areas. G. E. Leggett (*Wash. agric. Exp. Sta.*, 1959, Bull. 609, 16 pp.).—In 90 tests over 4 years wheat yields were highly correlated with available soil moisture in spring + rainfall during the growing season. Water required to grow the crop to the heading stage was 4 in., and yields were increased by 6 bushels per acre for each additional in. of water. In 62 tests yields were highly correlated with soil NO_3^--N content at sowing time + fertiliser N applied. Where N was limiting, yields were increased by 1 bushel per acre for each 3 lb. of N applied per acre.
A. H. CORNFIELD.

Fertilisation of dry-land wheat in Eastern Washington. G. E. Leggett, H. M. Reisenauer and W. L. Nelson (*Wash. agric. Exp. Sta.*, 1959, Bull. 602, 22 pp.).—In 112 tests over 5 years optimum N rate for wheat on summer-fallowed land ranged from 20—40 lb. per acre where rainfall was <10 in. to 30—80 lb. per acre where rainfall was >15 in. per annum. Anhyd. NH_3, aq. NH_3, $(NH_4)_2SO_4$, NH_4NO_3, $Ca(NO_3)_2$ and urea were equally effective as sources of N. Spring application of anhyd. or aq. NH_3 to winter wheat were as effective as autumn applications made before sowing. P applications had no effects on yields. A. H. CORNFIELD.

Grain drying and conditioning investigations. G. L. Nelson, G. W. A. Mahoney and J. I. Fryrear (*Okla. agric. Exp. Sta.*, 1959, Bull. 520, 30 pp.).—Field and laboratory drying trials of grain sorghum, wheat and rye using forced-air ventilation and natural wind ventilation are described. Trials with $NaHCO_3$ and two commercial materials, sold as powder admixtures to reduce damage during storage of grain containing excess moisture, were ineffective in preventing heating and loss of viability during storage of sorghum.
A. H. CORNFIELD.

Effects of basic tillage methods and soil compaction on maize production. H. P. Bateman (*Ill. agric. Exp. Sta.*, 1959, Bull. 645, 35 pp.).—The effects of various types of cultural implements, systems of cultural operations, fertiliser treatment and row spacing on the physical characteristics of the soil and on the crop response are examined. A. G. POLLARD.

Influence of magnesium on growth and development of maize inbreds and soya-bean varieties in nutrient solution culture and representative Ohio soils. R. D. Peel (*Dissert. Abstr.*, 1959, **20,** 1924—1927).—Strains of maize showing Mg-deficiency symptoms when grown in nutrient cultures containing 2 p.p.m. of Mg showed no Mg deficiency on 37 soils of low Mg content. Growth of maize and soya-beans on a soil with low exchangeable Mg was not improved by application of $MgSO_4$; Mg chlorosis was not induced by heavy applications of K. The strains of maize that showed Mg-deficiency symptoms on nutrient solutions with low Mg had lower contents of Mg than strains not showing these symptoms. Accumulation of ^{32}P was not correlated with the Mg content of the plant, or with Mg-deficiency symptoms. Varieties of soya-bean particularly sensitive to Mg deficiency were identified.
M. D. ANDERSON.

Responses of the rice plant to growth-regulating substances. A. J. Rampal and J. G. Bhatt (*Trop. Agric.*, 1959, **36,** 96—99).—Thirteen treatments are reported, but 2,3,5-triiodobenzoic acid alone promoted growth and yield of the rice plant to a certain extent. The others produced deleterious effects on plant development. There was no effect on earliness. E. G. BRICKELL.

Development of rice production and research in the Federation of Nigeria. J. E. Y. Hardcastle (*Trop. Agric.*, 1959, **36,** 79–95).—History and zones of cultivation, methods, varieties, processing, storage and consumption are reviewed together with current research on yield improvement, quality and soil fertility.
E. G. BRICKELL.

Seed germination and seedling growth of rice (*Oryza sativa*, L.) J. Dore and P. V. Thevan (*Trop. Agric.*, 1959, **36,** 15—34).—Germination of rice is adversely affected by covering the seed with soil, by stagnant (as opposed to running) water and by application of $(NH_4)_2SO_4$ immediately before sowing. These effects are associated with restricted aeration and/or accumulation of toxic gases in the soil. A. G. POLLARD.

Foliar sprays of urea and the rhizosphere microflora of rice. T. K. Ramachandra-Reddy (*Phytopath. Z.*, 1959, **36,** 286—289).—

Foliar applications of urea to rice seedlings stimulated the development of a *Penicillium* sp. which, in turn, diminished the population of bacteria and actinomycetes in the rhizosphere.
A. G. POLLARD.

Influence of grazing on plant succession of range lands. L. Ellison (*Bot. Rev.*, 1960, **26,** 1—78).—A review with an extensive bibliography. L. G. G. WARNE.

Fate of potassium applied to pasture on a soil derived from andesitic ash. W. M. H. Saunders and A. J. Metson (*N.Z. J. agric. Res.*, 1959, **2,** 1211—1231).—In pot experiments, addition of urine and KCl over a period of 6 months (spring to autumn) increased yields and K contents of grass and clover in undisturbed acres of a mixed pasture. The larger response of grass to urine was attributed mainly to N; the smaller response to KCl was entirely due to increased clover. Topdressing with KCl greatly increased the proportion of clovers. Leaching caused small losses of K, but losses of Mg indicated that continued dressing with KCl could lead to Mg deficiency. Without urine a deficiency level of K was reached within 3 months. (19 references.) K. R. BUTLIN.

Comparison of effects of ammoniacal and nitrate manuring on temporary grassland. P. Gouny, J. Picard, S. Mériaux and R. Grosman (*C. R. Acad. Agric. Fr.*, 1959, **45,** 889—893).—The first crops of an early grass obtained after N-manuring in Feb. show similar steady increases in yields of dry matter for increasing applications of either Nitro-chalk or $(NH_4)_2SO_4$; at 150 kg. of N per hectare, the latter gives 33% more sol. org. N, and 7% more protein in the grass than does the former. Considerable amounts of NH_4^+-N can, therefore, be absorbed before any appreciable nitrification can occur. As regards the second croppings (in May), the NO_3^--manured grass shows a positive, but the NH_4^+-manured grass a negative response to increasing N-manuring. Possible causes for these differences are considered. P. S. ARUP.

Nitrogen-manuring and yield of grassland. D. Oostendorp (*Landbouwvoorlichting*, 1960, **17,** 39—46).—The effects of annual N-manuring do not decline over periods of 10—20 years. Since yields of dry matter per kg. of N decrease (especially on peaty soil) with increasing applications, the rates should be kept within the range 100—200 kg. per hectare. Excessive N-manuring may cause digestive troubles in cattle due to excess of protein in the grass.
P. S. ARUP.

Mixtures of lucerne and grasses. S. Wlodarczyk (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E,** 173—196).—*Medicago sativa* in admixture with *Dactylis glomerata* L. *Festuca pratensis*, *Arrhenatherum elatius* and *Phleum pratense* was tested on a silty soil plus sand. Highest hay yield was from *Medicago sativa* and *Dactylis glomerata* and the lowest from lucerne in pure culture. In the fourth year of cropping the best yield was given by the mixture *Medicago sativa* and *Festuca pratensis*. No correlation existed between protein content, fibre and ash in hay of the first, second and third mowing. (15 references.) E. G. BRICKELL.

Winter growth of lucerne (*Medicago sativa*). I. Variety performance in New Zealand. T. P. Palmer (*N.Z. J. agric. Res.*, 1959, **2,** 1195—1202).—Several lucerne varieties covering a wide range of winter activity gave similar performances at five N.Z. stations in different years and in winter and spring. One suitable variety should give high winter production wherever lucerne is normally grown. The order of winter growth of different varieties was quite different from that at Cambridge, England. K. R. BUTLIN.

Cold hardiness in *Trifolium repens* and *Medicago sativa*, L. C. G. Greenham and H. Daday (*Aust. J. agric. Res.*, 1960, **11,** 1—15).—The cold resistance and/or cold injury in these species is measured by low (*L*)- and high (*H*)-frequency resistances or by the resistance index ($L/F \times 100$). A. G. POLLARD.

Occurrence of α- and β-carotenes in plants. IIA. Fodder plants: lupin. A. Sykut and Z. Wierzchowski (*Ann. Univ. M. Curie-Sklodowska*, 1958 [1959], **13C,** 261—274).—The max. total carotene concn. (573 mg./kg. dry wt.) occurs in the leaves at the beginning of the flowering period and during formation of the first pods. The average content of the α-isomer is small (8·4%). E. M. J.

Effect on beet seed of low germination temperatures. J. Séchet (*C. R. Acad. Agric. Fr.*, 1959, **45,** 879—881).—A marked growth response is obtained by keeping the seed, previously germinated during 4 days at 23°, at 2° during 57 or (preferably) 45 days before transplantation. The economically useful material obtained from the treated seeds is approx. double that obtained from untreated seeds transplanted at the same stage of germination.
P. S. ARUP.

Productive vernalisation of sugar and forage beets. S. Lewicki and B. Mysakowska-Paleolog (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E,** 361—377).—Under favourable atm. conditions, particularly in regard to rainfall, vernalisation can have a positive influence on root yields, +3° for 3 days giving max. results. No influence was detected on leaf yields nor on the content of dry matter or sugar in the roots. E. G. BRICKELL.

Influence of leaf-cutting terms on yield and chemical composition of chicory (*Cichorium intybus*, L.). S. Tabin and J. Plotnicki (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E,** 379—401).—In two varieties the dry leaf mass was lower with one term of leaf cutting than with two but the yields of nutritive constituents in the leaves were much higher with repeated leaf cutting. Chemical composition of roots was unaffected but repeated leaf cutting decreased the root yield by about 30%. (22 references.) (From English summary.)
E. G. BRICKELL.

Irrigation experiments with cotton, maize, sorghum and castor beans. J. F. Garton and A. D. Barefoot (*Okla. agric. Exp. Sta.*, 1959, Bull. 534, 19 pp.).—During dry years cotton yields were highest per acre and per acre-in. water supplied with the highest level of irrigation. During wet years yields per acre were also highest at the highest irrigation level, but yields per acre-in. of water supplied were highest at the lowest level of irrigation. High levels of irrigation water were not economical for sorghum, and temporary moisture stresses did not decrease yields of this crop. Maize gave the highest yields per acre and per acre-in. of water applied at the highest level of irrigation. Castor beans gave highest yields with the high level, but the highest yields per acre-in. with the low level of irrigation water. A. H. CORNFIELD.

Bitter pit of apples. I. Physical and chemical changes in leaves and fruits of Cox's Orange variety during the season. H. O. Askew, E. T. Chittenden, R. J. Monk and J. Watson (*N.Z. J. agric. Res.*, 1959, **2,** 1167—1186).—Data for fruits and leaves of Cox's Orange variety during 1957—58 and 1958—59 are presented. The developing fruits showed decreases in mineral concn. and increases in sugar as they matured. The ratio Ca/Mg, perhaps important in governing incidence of bitter pit, varied considerably with growth. Fresh leaves were much richer than fruits in minerals and N.
K. R. BUTLIN.

Periodicity in the growth of fruits of apples, peaches and sour cherries with some factors influencing this development. L. D. Turkey (*Pa agric. Exp. Sta.*, 1959, Bull. 661, 21 pp.).—The relationship between the rhythm of growth of the fruit of apple, peach and sour cherry and other factors (moisture stress, climatic conditions, variety) is reported. A. H. CORNFIELD.

Characteristics of citrus fruits on different rootstocks. A. Wallace (*Univ. Cal. Los Angeles*, Spec. Rep., 1958, No. 1, 125—128).—Various citrus fruits on rough lemon rootstocks or, occasionally, on sown orange stock showed lower % of juice and wt. of acid and sol. solids per unit wt. of fresh fruit than when other rootstocks were used. A. G. POLLARD.

Influence of leaf urea sprays on leaf-burn and on leaf, stem and root yields and nitrogen contents of citrus cuttings. J. R. Kuykendall and A. Wallace (*Univ. Cal. Los Angeles Spec. Rep.*, 1958, No. 1, 162—173).—Spraying the leaves with urea (1—4%) increased the N content of leaves, stems and roots, increased yields when the root supply of N was low and slightly lowered yields when the N supply was high. Addition of sugar to the spray diminished the N absorption and the incidence of leaf-burn. Girdling did not affect absorption of urea from the spray but tended to reduce leaf-burn from the more conc. sprays. A. G. POLLARD.

Variability in nitrogen content of lemon leaves of the same age. T. A. Mouloulis and A. Wallace (*Univ. Cal. Los Angeles, Spec. Rep.*, 1958, No. 1, 155—162).—For assessing the nutrient of lemon trees 9—15 leaves form an adequate sample to give ±5% accuracy. The N content of leaves of fruiting shoots or shaded shoots was below that of other shoots. No translocation of N occurred from leaves <6 months old to zones of newer growth.
A. G. POLLARD.

Deficiency symptoms of the major elements in the banana. D. B. Murray (*Trop. Agric.*, 1959, **36,** 100—107).—Deficiency symptoms produced by N, P, K, Ca and Mg are described and illustrated.
E. G. BRICKELL.

Pruning bananas with 2,4-D. J. O. Wright (*Trop. Agric.*, 1959, **36,** 285).—Treating unwanted suckers with about 5 ml. of 2% aq. Na salt applied from a plastic wash-bottle into the small cavity formed by the base of a petiole, or into the centre of the rolled-up spear, proved effective. E. G. BRICKELL.

Influence of growth-promoting substances on pineapples. H. R. Evans (*Trop. Agric.*, 1959, **36,** 108—117).—1-Naphthylacetic acid, Hortomone A, Seradix A and Planofix all increased fruiting and accelerated harvesting. Dicotox suppressed fruiting and harvesting but fruit wt. was increased. Anapal and Tomato Set also suppressed fruiting. E. G. BRICKELL.

Zinc deficiency in cacao in Netherlands New Guinea. H. Schroo (*Neth. J. agric. Sci.*, 1959, **7**, 309—316).—Acute leaf symptoms of Zn-deficiency occurred on young cacao trees growing on an acid leached quartzitic silt loam and on an alluvial fan silt loam (containing free $MgCO_3$). Spraying the trees every 10 days with aq. $ZnSO_4$ (2·5 g. per tree) for 6 months resulted in almost complete elimination of Zn-deficiency symptoms and much increased growth in comparison with controls. A. H. CORNFIELD.

Pest Control

Microbial insecticides and their future. G. Decker (*Agric. Chemic.*, 1960, **15**, No. 1, 30—33, 93).—A general account of the advantages and disadvantages of biological methods of pest control. A. H. CORNFIELD.

Designation of chemicals in terms of the responses they elicit from insects. V. G. Dethier, L. B. Browne and C. N. Smith (*J. econ. Ent.*, 1960, **53**, 134—136).—As the terms attractant and repellant are commonly used to cover a wide range of reactions, six terms are defined to cover all aspects. (13 references.) C. M. HARDWICK.

DDT-dehydrochlorinase. III. Solubilisation of insecticides by lipoprotein. H. Lipke and C. W. Kearns (*J. econ. Ent.*, 1960, **53**, 31—35).—The solubility data for various insecticides in egg yolk lipoprotein are given; these are 10^4 or 10^5 times greater than in water. The use of this substrate in studies of enzyme kinetics is discussed. (26 references.) C. M. HARDWICK.

Laboratory method for screening compounds as attractants to gypsy moth males. B. C. Block (*J. econ. Ent.*, 1960, **57**, 172—173).—Male *Porthetria dispar*, secured by spring type clothes pin mounts, were exposed to various compounds and the occurrence of the copulatory behaviour pattern recorded. Benzene, acetone, octyl caprylate and 1,2-epoxyhexadecane produced the reactions expected from field tests. C. M. HARDWICK.

Effects of spray mixtures on toxicity of DDT, parathion and malathion. W. S. Hough, A. B. Groves and C. H. Hill (*Virginia agric. Exp. Sta.*, 1959, Tech. Bull. 143, 26 pp.).—The presence of S in the spray improved the residual toxicity of DDT against the codling moth larva, whilst lime-S reduced the toxicity of DDT in most tests. The presence of Bordeaux mixture improved the residual but not the initial toxicity, whilst ferbam and zineb had no effect on toxicity of DDT. The residual toxicity of parathion was not affected by the presence of fungicides. Initial toxicity was reduced by the presence of S fungicides, with the exception of lime-S. The residual toxicity of malathion was not affected by the presence of fungicides. A. H. CORNFIELD.

Effect of carbon disulphide fumigation on *Trichoderma viride* and other soil fungi. S. B. Saksena (*Trans. Brit. mycol. Soc.*, 1960, **43**, 111—116).—After fumigation with CS_2, *T. viride* became the dominant soil fungus when the dosage was between 0·05 and 1·0 ml. of CS_2 per 250 g. of soil. The tolerance of *T. viride* to CS_2 was lower than that of several species of *Penicillium* and *Aspergillus*. L. G. G. WARNE.

Persistence of DDT, aldrin and lindane in some mid-western soils. E. P. Lichtenstein, L. J. DePew, E. L. Eshbaugh and J. P. Sleesman (*J. econ. Ent.*, 1960, **53**, 136—144).—Various soils were treated with each insecticide at three dosages. The residues were measured colorimetrically and by bioassay with *Drosophila melanogaster* over 4½ years. The greatest decline was in the first half year; DDT persisted longer than lindane or aldrin. Persistence was greater in soils with a higher level of org. matter and in dry soils. (15 references.) C. M. HARDWICK.

Fate of Phorate in soils. L. W. Getzin and R. K. Chapman (*J. econ. Ent.*, 1960, **53**, 47—51).—Cabbage, potato and pea plants took up more Phorate (**I**) (Thimet) from sand or sandy soil than from loams or muck soils, as shown by anticholinesterase assay. Leaching from the top 1½ in. varied in 24 days from 75% in quartz sand to 7% in muck. Over 4 weeks the % of **I** in soil decreased and its oxidation products increased. The amount of "bound" **I** was proportional to the org. content of a soil. Volatilisation of **I** from soil was significant for only 1 hr., but for 24 hr. in quartz sand. C. M. HARDWICK.

BHC translocation from treated soil and the effect on growth of red clover. C. L. Brass and G. W. Ware (*J. econ. Ent.*, 1960, **53**, 110—113).—Clover grown on a clay and a silt loam treated with BHC (0·1, 1·0 and 100 p.p.m.) was analysed. Amounts in that grown on clay loam were 10—20% lower. There was no reduction in germination although some plants were malformed. All plants surviving until the 9-week stage recovered. C. M. HARDWICK.

Germination of cereal sorghum and small legume seeds after fumigation with hydrogen phosphide. R. G. Strong and D. L. Lindgren (*J. econ. Ent.*, 1960, **53**, 1—4).—The germination of seeds of barley, maize, oats, rice, wheat, milo grass, lucerne, clover and trefoil with moisture content 9—15% was unaffected by fumigation with up to 18 mg./l. of hydrogen phosphide for 7 days at 50—90°F. C. M. HARDWICK.

***O*-Ethyl *S*-2-(ethylthio)ethyl alkylphosphonothioates as systemic insecticides.** R. L. Metcalf and T. R. Fukuto (*J. econ. Ent.*, 1960, **53**, 127—130).—The properties of 10 of the compounds are tabulated. The results obtained for the LD_{50} when topically applied to houseflies, bioassay of isolated leaves from treated cotton plants and treated seeds could be predicted from their structure. (13 references.) C. M. HARDWICK.

Relationship between metabolism and differential toxicity in insects and mice of diazinon, Dimethoate, parathion and Acethion. H. R. Krueger, R. D. O'Brien and W. C. Dauterman (*J. econ. Ent.*, 1960, **53**, 25—31).—Labelled insecticides were injected into mice and *Periplaneta americana* and applied topically to houseflies and the results used to explain the selective nature of their toxicity. (12 references.) C. M. HARDWICK.

Comparison of toxicity of insecticides for control of maize earworm on sweet maize. L. D. Anderson and H. T. Reynolds (*J. econ. Ent.*, 1960, **53**, 22—24).—Of 69 compounds brushed on to individual maize ears, only Sevin, Thiodan, heptachlor, Dipterex and isodrin gave equal or better control of *Heliothis zea* than did 5% DDT. Higher doses of malathion, diazinon and Guthion were also effective. C. M. HARDWICK.

Control of sap beetles in sweet maize grown for processing. F. P. Harrison (*J. econ. Ent.*, 1960, **53**, 174—175).—In field experiments the addition of malathion and Dylox to DDT sprays for maize earworm control considerably reduced the number of ears infested with *Carpophilus lugubris*. C. M. HARDWICK.

Corky ringspot of potatoes, a soil-borne virus disease. C. H. Walkinshaw and R. H. Larson (*Wis. agric. Exp. Sta.*, Res. Bull. 217, 31 pp.).—Corky ringspot of potatoes is due to a complex culture composed of two viruses (etching and necrotic isolates). The reactions of 29 species to sap inoculation were similar to those induced by inoculation with tobacco rattle virus. Infested soil could act as a source of primary inoculum and a no. of solanaceous species were susceptible to virus invasion from the soil. *Vinca rosea* was very suitable for maintaining stock cultures of the viruses. Samsum tobacco was particularly susceptible to infection from soil as well as from mechanical inoculation. A. H. CORNFIELD.

Black beetle (*Heteronychus sanctae-helenae* Blanch.) in pastures in New Zealand. D. H. Todd (*N.Z. J. agric. Res.*, 1959, **2**, 1262—1273).—The seasonal history, habits, distribution and general ecology of the beetle in N.Z. pastures are described. It has only one generation per year. Both larvae and adults damage pastures, causing continuous deterioration and hence lower stock-supporting capacity. Dieldrin (4, preferably 8, oz./acre) effectively controls the beetle in pastures. K. R. BUTLIN.

Control of white grubs, *Eulepida mashona*, Arrow. C. E. Taylor and C. N. Smithers (*Rhod. agric. J.*, 1959, **56**, 240—242).—Application of dieldrin (1 lb. per acre) to a grass sward gave almost complete control of whitegrubs and greatly improved coverage of grass. A. H. CORNFIELD.

Effect of Dylox and other insecticides on entomophagous insects attacking field crop pests in California. V. M. Stern, R. van den Bosch and H. T. Reynolds (*J. econ. Ent.*, 1960, **53**, 67—72).—The effect of sprays of Dylox, DDT, toxaphene, parathion, Sevin and heptachlor on beneficial species of insects on lucerne, is listed. At $<\frac{1}{2}$ lb./acre, Dylox was only slightly to moderately toxic to coccinellid adults and larvae, *Chrysopa* spp. and *Geocoris* spp. It is relatively non-toxic to bees. Its short residual effect is also favourable to insect survival. C. M. HARDWICK.

Potential use of Dylox and other insecticides in a control programme for field crop pests in California. H. T. Reynolds, V. M. Stern, T. R. Fukuto and G. D. Peterson, jun. (*J. econ. Ent.*, 1960, **53**, 72—78).—Dylox was no more effective than other compounds against lepidopterous larvae attacking lucerne hay, *Lygus* and related species attacking lucerne seed and hemiptera and lepidoptera attacking cotton. The short residual activity made it less harmful to beneficial insects. At rates of 1 lb./acre residues on lucerne hay were <1 p.p.m. after 4 days. C. M. HARDWICK.

Biology and control of apple seed chalcid in North Carolina. G. F. Turnipseed (*J. econ. Ent.*, 1960, **53**, 166).—*Torymus druparium* larvae overwintered for one or two seasons in the apple seed, pupated in late April and emerged in May. One spray of DDT, malathion or Sevin in May gave effective control. Demeton, Thimet and Delnav were ineffective. C. M. HARDWICK.

Response of different strains of green peach aphid to malathion. F. H. Shirck (*J. econ. Ent.*, 1960, **53**, 84—88).—Comparison of different strains did not show clear-cut differences in tolerance to

malathion but some did show increased tolerance over several generations. In four tests aphids with wing pads were harder to kill. C. M. HARDWICK.

Soil injection as a means of applying systemic acaricides to fruit trees. M. L. Cleveland (*J. econ. Ent.*, 1960, **53,** 144—146).—An air-pressure and a water pressure applicator are described. Injection of Phorate reduced no. of *Panonychus ulmi* greatly in two orchards but Chipman R-6199 was less effective. C. M. HARDWICK.

Control of sugar-beet wireworm in southern California bean fields. M. W. Stone and L. D. Anderson (*J. econ. Ent.*, 1960, **53,** 176—177).—Experiments in two fields over 6 years for control of *Limonius californicus* showed that sprays of aldrin, toxaphene, DDT + toxaphene were satisfactory. C. M. HARDWICK.

Control of root maggots in swede turnips in Newfoundland with heptachlor and aldrin and the effect on parasites and overwintering pupae. R. F. Morris (*J. econ. Ent.*, 1960, **53,** 65—67).—Application of 2 and 4 lb./acre of granulated heptachlor was more effective than equivalent doses of aldrin in controlling *Hylemya* spp. All treatments more than doubled the number of marketable roots. The numbering of overwintering puparia was also correlated with the effectiveness of control. C. M. HARDWICK.

Nicotine residues in field-treated cauliflower, celery, green beans, kale, mustard greens and spinach. L. D. Anderson and F. A. Gunther (*J. econ. Ent.*, 1960, **53,** 64—65).—The use of Black Leaf-40 at normal doses left no appreciable residues with green beans, celery and cauliflower curds. Kale and spinach retained residues for one week and Texas mustard greens for two weeks.
C M. HARDWICK.

Control of the bean stem maggot, *Agromyza phaseoli*, Coq. C. E. Taylor (*Rhod. agric. J.*, 1959, **56,** 195—196).—Treatment of bean seed with 0·2% aldrin, dieldrin or endrin gave excellent protection against the bean stem maggot and had no phytocidal effects.
A. H. CORNFIELD.

Onion diseases and their control. J. C. Walker (*U.S. Dep. Agric.*, 1959, Fmrs Bull. 1060, 26 pp.).—Fourteen diseases primarily important in the field and seven diseases important in storage and transit are described. Onion smut may be controlled by formaldehyde drip or thiram dust applied to the seed but most of the other diseases respond only to crop rotation, timely cultivation, use of resistant varieties and disease-free bulbs, sets and seed, sanitation and steam sterilisation. Storage is best at just above 32°F and 70—75% R.H.
E. G. BRICKELL.

Biogenesis of fusaric acid. R. S. Sandhu (*Phytopath. Z.*, 1959, **37,** 33—60).—Production of fusaric acid by *Fusarium lycopersici* in culture media and the effects thereon of various amino-acids added to the medium, are examined. Glucose is utilised in the formation of the acid, the precursors of which include α-alanine, serine and acetic and γ-aminobutyric acids. Production of the acid by *Gibberella fujikuroi* was also examined. A. G. POLLARD.

Action of fusaric acid on polyphenol-oxidase. R. Bossi (*Phytopath. Z.*, 1960, **37,** 273—316).—Fusaric acid inhibits the activity of polyphenol-oxidase and also, at much higher concn., that of catalase and ascorbic acid oxidase. These effects are discussed in relation to the development of tomato wilt disease. A. G. POLLARD.

Control of the Fuller Rose beetle on citrus in California. H. S. Elmer (*J. econ. Ent.*, 1960, **53,** 164—165).—Sprays of endrin, aldrin, dieldrin, chlordane, toxaphene and cryolite reduced the no. of *Pantomorus godmani* considerably. In laboratory tests the duration of protection of foliage from adult feeding by 16 compounds was found. C. M. HARDWICK.

Cotton insects. J. C. Gaines (*Texas agric. Exp. Sta.*, 1959, Bull. 933, 16 pp.).—Harmful and beneficial insects are described. In some areas cotton insect pests have developed resistance to chlorinated hydrocarbons. A. H. CORNFIELD.

Seasonal study of diapause, reproductive activity and seasonal tolerance to insecticides in the boll weevil. J. R. Brazzel and B. G. Hightower (*J. econ. Ent.*, 1960, **53,** 41—46).—The reproductive activity of *Anthonomus grandis* occurred when the cotton was growing actively. Diapause started in July/Aug. but renewed growth in Sept. could reverse the trend. Diapausing weevils with a fat content >10% were not susceptible to chlorinated hydrocarbons but reproducing weevils were as much as 100 times more susceptible.
C. M. HARDWICK.

Increased tolerance of the boll weevil and cotton fleahopper to some chlorinated hydrocarbon insecticides in central Texas in 1958. C. R. Parencia, jun. and C. B. Cowan, jun. (*J. econ. Ent.*, 1960, **53,** 52—56).—In seven field experiments *Anthonomus grandis* was resistant to chlorinated hydrocarbons although toxaphene + DDT gave better results than did other treatments. Early season populations of *Psallus seriatus* also survived chlorinated hydrocarbons. Sevin and Guthion gave satisfactory control of both weevils.
C. M. HARDWICK.

Salt-marsh caterpillar control on cotton in Arizona. G. P. Wene, D. M. Tuttle and L. W. Sheets (*J. econ. Ent.*, 1960, **53,** 78—80).—In 1955, good control of *Estigmene acrea* was obtained with sprays containing parathion + toxaphene and dusts of DDT + toxaphene. In 1958, DDT + toxaphene gave only 50% control while Dilan and parathion gave >97% control. C. M. HARDWICK.

Laboratory studies on the effectiveness of Chipman-6199 against some cotton pests. C. H. Tsao and G. T. Bottger (*J. econ. Ent.*, 1960, **53,** 103—106).—Sprays of Chipman R-6199 [the monohydrogen oxalate of *OO*-diethyl *S*-(2-diethylamino)ethyl phosphorothioate] (**I**) on cotton plants were similar in toxicity to those of endrin to lepidopterous larvae, except the bollworm, to which it is less toxic. With 5 mg. per plant good control was given of *Tetranychus telarius* and *Hercothrips phaseoli* for 48 days; *Aphis gossypii* for 25 days. Adult *Anthonomus grandis* and *Pectinophora gossypiella* and *Coccus hesperidus* were controlled for only 2 days. When **I** was mixed with DDT, the toxic action was additive. The greatest absorption occurred in the first 2 days but detached leaves continued absorption for a week. Translocation occurred from the lower leaves to new ones. C. M. HARDWICK.

A seedling die-back of *Theobroma cacao*, L. in Nigeria. I. Description of the disease and its spread in the nursery. S. R. Chant. **II. Factors affecting the incidence of the disease and its control.** S. R. Chant and T. H. R. Hall (*Trop. Agric.*, 1959, **36,** 138—144, 145—149).—I. Symptoms of the disease caused by *Phytophthora* sp. are described. It appears in the first instance to occur at random in the nursery; subsequent spread is from infected to adjacent plants.
II. Heavy shade and high R.H. favour the disease and the incidence of die-back was greater in a heavy soil than in a sandy one. Soil sterilisation and seed dressing had no effect on the subsequent incidence but applications of Perenox spray (0·3 and 0·5%) at 3- or 6-day intervals from 3 weeks after sowing gave control.
E. G. BRICKELL.

Increased cocoa production by foliar copper applications as an effect additional to Witches' Broom disease control. D. Tollenaar (*Trop. Agric.*, 1959, **36,** 177—188).—Mist-blown applications of Copper-Sandoz considerably reduced the no. of diseased pods and vegetative brooms. Dithane was not effective.
E. G. BRICKELL.

Control of oak wilt disease. E. B. Himelick (*Dissert. Abstr.*, 1959, **20,** 1554—1555).—The loss of marketable oak timber caused by the oak wilt fungus, *Ceratocystis fagacearum*, in N. Illinois was reduced by poisoning infected trees, and also healthy trees within root-grafting distance of infected trees, and establishing poison barriers round centres of infection. Na arsenite killed trees more rapidly than any other substance tested, and also killed roots to some extent. Vapam and methyl bromide applied to the soil killed roots up to 3 in. in diameter, and to a depth of 3 ft. The oak wilt fungus can be used to destroy unwanted oak brush in pine plantations. M. D. ANDERSON.

Toxicity of BHC to loblolly pine seedlings. R. C. Thatcher (*J. econ. Ent.*, 1960, **53,** 175—176).—Total immersion of pine seedlings in 1 or 2% BHC produced stunted growth, and high mortality at the 2% dosage. Top dips were not phytotoxic and gave good weevil control. Water emulsions and suspensions produced similar results. C. M. HARDWICK.

Control of stem rot of groundnuts. K. H. Garren (*Va agric. Exp. Sta.*, 1959, Tech. Bull. 144, 29 pp.).—Characteristics of the disease, due to *Sclerotium rolfsii*, are described. Deep covering of org. matter during seed-bed cultivation combined with careful cultivation so as to avoid dirtying the stems reduced infection, increased pod yields, and improved the quality of the nuts. A. H. CORNFIELD.

Effect of Phorate applied to seed on the growth, development and insects attacking grain sorghum. R. T. Everly and R. C. Piquett (*J. econ. Ent.*, 1960, **53,** 154—160).—With Phorate at 2 and 4 lb./acre, stands were reduced considerably but were 10% better when Arasan was included. Some reduction in germination after storage may be due to the sticker. Treatment lengthened the time from germination to plant emergence and, in general, reduced yields. Higher doses of Phorate without Arasan reduced no. of *Rhopalosiphum maidis* by the greatest amount. The effectiveness of seed treatment was reduced as the plant grew. (13 references.)
C. M. HARDWICK.

Effect of methyl bromide and hydrocyanic acid fumigation on germination of rice. R. G. Strong and D. L. Lindgren (*J. econ. Ent.*, 1959, **52,** 706—710).—Fumigation with HCN had no effect on rice germination. A reduction in % germination following methyl bromide fumigation occurred with increases in moisture content, time

of exposure and temp. of 50—70°F and (to a smaller extent) of 70—90°F. These factors were interrelated. Results were similar 5 and 84 days after treatment. C. M. HARDWICK.

Effect of methyl bromide and hydrocyanic acid fumigation on germination of flax seeds. R. G. Strong and D. L. Lindgren (*J. econ. Ent.*, 1960, **53,** 17—19).—No reduction in germination resulted from exposure to HCN for 72 hr. at temp. up to 90°F of flax seeds with 6—12% moisture content. Erratic damage occurred at different temp., lengths of exposure moisture contents and with a second fumigation when methyl bromide was used. Only varietal differences and post-fumigation storage time were not significant. C. M. HARDWICK.

Control of thread blight disease, *Pellicularia koleroga*, Cooke, of pepper. T. Theis, L. Calpouzos, L. Gregory and N. Almeyada (*Plant. Prot. Bull.*, 1959, **7,** 161—162).—The disease was controlled by two applications of Bordeaux mixture (5–5–50) or "Copper A" at 3-week intervals. A. H. CORNFIELD.

Effect of insecticides on transamination in the American cockroach J. W. McAllan and A. W. A. Brown (*J. econ. Ent.*, 1960, **53,** 166—167).—As Na orthoarsenate, DDT, malathion, toxaphene and endrin did not inhibit the glutamate–aspartate transamination when injected into cockroaches, it is suggested that the *in vitro* inhibition is an artifact. C. M. HARDWICK.

Weed infestation of fields at Slawin with special reference to content of seeds in soil. W. Kulpa and F. Pawlowski (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E,** 243—300).—The no. of weeds per sq. in. varied from 148 to 1018 individuals and the no. of species from 20 to 39. The total no. of weed seeds in 1 sq. in. of arable layer (20 cm. deep) varied between approx. 16,000 and 44,300, the no. of species between 30 and 45. The no. of perennial species was greatest in clover, i.e., in a crop which dispenses with ploughing and decreases in annual cultures. The reverse followed for annual weeds. (27 references.) (From English summary.) E. G. BRICKELL.

Analysis of substituted acetic acids used as herbicides. IV. Separation of trichloroacetic acid and 2,4-dichlorophenoxyacetic acid. V. Joint determination of 2,4-dichlorophenoxyacetic acid and 2,4,5-trichlorophenoxyacetic acid by differential-solubility method. B. T. Milićević and (IV) S. D. Janković, (V) I. L. Kostić (*Bull. Soc. chim. Belgrade*, 1958/59, **23—24,** 67—73, 75—77).—IV. Three procedures are given for the separation of the two acids by (*i*) ascending paper-strip chromatography, (*ii*) countercurrent distribution in a 25-tube apparatus, and (*iii*) decomposition of trichloroacetic acid with NaOH. (18 references.)

V. A mixture of the two acids dissolved in acetone is titrated with water to the first appearance of turbidity. The log. curve for ml. of water *vs.* acid concn. is linear; max. error is 2%. W. J. BAKER.

Trials of herbicides in banana plantations. J. Champion and J. Monnet (*Fruits d'outre mer*, 1959, **14,** 459—464).—The dwarf banana in French Guinea yields more heavily if the soil between the trees is kept free of vegetation. In preliminary tests in French Guinea, monuron checked the growth of weeds for a period depending on the rainfall after application, and was not toxic to the bananas if it did not fall on their leaves. In Guadaloupe, urea herbicides mixed with contact herbicides and applied in an oil spray gave promising results. M. D. ANDERSON.

2,4-D injury to crops and ornamental plants. M. B. Linn, F. W. Slife and B. J. Butler (*Ill. agric. Exp. Sta.*, 1959, Circ. 808, 15 pp.).—The symptoms and extent of injury by 2,4-D are described and the influence of method of formulation (high- and low-volatile esters, amines) and type of spray jet is examined. Means of preventing or minimising spray injury are noted. A. G. POLLARD.

Effect on groundnuts (*Arachis hypogea*) of [A] pre-emergence, [B] post-emergence applications of MCPB. P. H. Rosher and R. D. Sheldrick (*Trop. Agric.*, 1059, **36,** 118—129, 211—217).—[A] 4-(4-Chloro-2-methylphenoxy)butyric acid (**I**) can be applied successfully at 2 lb./acre 3 days after sowing. A list is given of the weed species likely to be controlled or suppressed by this treatment.

[B] **I** at 0·94 lb./acre can be applied safely 1 or 2 weeks after emergence or between 2·25 and 5·53 lb./acre 3 weeks after emergence. In the field 1·0 lb./acre applied 1 or 2 weeks after emergence does not depress the yield of groundnuts that are kept clean by hand weeding and 4·0 lb./acre applied 3 weeks after emergence also had no effect on yield; 6·0 lb./acre at 3 weeks after planting however caused a serious reduction in yield. E. G. BRICKELL.

Control of mesquite, *Prosopis juliflora*, in grazing lands. C. E. Fisher, C. H. Meadors, R. Behrens, E. D. Robinson, P. T. Marion and H. L. Morton (*Texas agric. Exp. Sta.*, 1959, Bull. 935, 24 pp.).—Cultural and chemical methods of control, including aircraft application, of 2,4,5-T are described. A. H. CORNFIELD.

Chemical eradication of woody plants. C. B. Owens (*Dissert. Abstr.*, 1959, **20,** 1922—1924).—Spraying with polypropylene glycol butyl esters of 2,4-D and 2,4,5-T in diesel oil was studied. When hawthorn was treated in Feb.–May, small amounts of herbicide in large vol. were more effective than the same or larger amounts in smaller vol. of oil. The junction of root and trunk was the most effective point of application. Spraying the foliage killed only the leaves treated. Hawthorn stumps were killed by spraying. *Ailanthus* was killed by spraying the lower part of the trunk in March; with low, but not with high, concn. of herbicide, sprouting occurred from the roots. Willows sprayed in May were top-killed in all cases; extensive sprouting occurred from the roots when the root crowns were under water, but not when they were above water. Japanese honeysuckle sprayed in March was top-killed, but sprouted when herbicide concn. was low. A graduated attachment for a 3-gal. knapsack sprayer was devised, to measure small amounts of spray accurately. M. D. ANDERSON.

Effects of arboricides on East African trees and shrubs. II [A] Species of the *Brachystegia-Pseudoberlinia* woodland. G. W. Ivens (*Trop. Agric.*, 1959, **36,** 52—64).—Spraying the frilled bases of these species with 2,4,5-T (ester formulation) killed >90% of the above-ground parts of the trees. 2,4-D, alone or in admixture with 2,4,5-T, was less effective but all chemicals gave better results than did ring-barking. Oil solutions of 2,4,5-T were superior to emulsions. Frilling of the trunks was essential. A. G. POLLARD.

Effects of arboricides on East African trees and shrubs. II. [B] *Combretum* and *Commiphora* species. G. W. Ivens (*Trop. Agric.*, 1959, **36,** 219—229).—NH_4 sulphamate was ineffective in aq. solution as a basal spray or applied in the form of crystals to stumps. Monuron showed promise when applied at a high rate. 2,4,5-T at 1·5—2·0% and also 2,4-D were effective particularly on *Combretum* species. Frilling generally increased the effectiveness of treatment. Little difference between treatments applied at different times of year was noted with *Combretum* species, but with *Commiphora schimperi* a slightly greater development of regeneration took place after treatment in the dry season than during the rains. E. G. BRICKELL.

Food reserves in post oak, *Quercus stella*, stumps and roots. W. C. Elder and J. E. Webster (*Okla. agric. Exp. Sta.*, 1959, Tech. Bull. 80, 11 pp.).—Chemical analysis of borings of root and trunk taken 10—12 in. above ground level were made over 5 years on (*a*) uncut trees, (*b*) trees cut 12—18 in. above ground and (*c*) trees cut and the stumps treated with 2,4-D + 2,4,5-T in diesel oil. Sol. solids and sugars were somewhat lower in treated stumps than in untreated stumps and uncut trees. There were little differences in total and acid-hydrolysable N between the treatments. Roots contained more sugars and total N than did stumps. Highest food reserves occurred in Dec./Jan. and lowest in May. Results are discussed in relation to time of treatment of stumps with hormone killers. A. H. CORNFIELD.

Thiophosphoric acid esters. Farbenfabriken Bayer A.-G. (B.P. 808,879, 4.7.57. Ger., 10.7.56).—Compounds $1,4,2\text{-}SO_nR\cdot C_6H_3R'\cdot CH_2\cdot S\cdot PX(OR'')\cdot OR'''$, useful as insecticides and plant-protectants, are obtained by interaction of $1,4,2\text{-}SO_nR\cdot C_6H_3R'\cdot CH_2Y$ with a salt of $OR''(OR''')\cdot PX\cdot SH$ at 20—80° in a solvent, e.g., low-mol. alkanol, aliphatic ketone, or aromatic hydrocarbon (*n* is 0—2, X is O or S, Y is halogen; R—R''' are alkyl of 1—4 C). The prep. is detailed for OO-*diethyl* S-2-*methylthio*-5-*methylbenzyl phosphorothiolate*, b.p. 144°/0·01 mm., which is 100% lethal to spider mites at a concn. of 0·01%. F. R. BASFORD.

Thiophosphoric acid esters. Farbenfabriken Bayer A.-G. (B.P. 811,677, 6.9.56. Ger., 7.9.55).—Compounds $(OR)_2PX\cdot S\cdot Y\cdot CO\cdot NH\cdot COR'$ (R is alkyl of 1—4 C; X is O or S; Y is alkylene of 1—4 C; R' is alkoxy or aryloxy), useful as pesticides are obtained by interaction of a salt of $(OR)_2PX\cdot SH$ with a halogenoacyl-urethane in an inert solvent. OO-*Diethyl* S-(N-*ethoxycarbonylcarbamoyl*)*methyl thiothionophosphate*, m.p. 73°, is prepared. F. R. BASFORD.

Amidic esters of dithiophosphoric acids and insecticidal compositions including them. Montecatini Società Generale per l'Industria Mineraria & Chimica (B.P. 808,853, 15.3.56. It., 25.3.55).—Compounds $(OR)_2PS\cdot S\cdot CHR'\cdot CO\cdot NR''R'''$ and insecticidal compositions containing them are claimed (R is alkyl; R' is alkyl or aryl; R'' and R''' are H or alkyl). As an example of the method of prep.,

that of OO-*diethyl* S-(2-iso*propylcarbamylethyl*) *thiothionophosphate*, m.p. 41—43°, is detailed. Topical application of this in acetone to female flies is 100% lethal at a dose of 2 μg. per fly.
F. R. BASFORD.

Sulphinyl-phosphoric acid esters. Farbenfabriken Bayer A.-G. (Inventor: G. Schrader) (B.P. 811,268, 4.12.57).—Sulphinyl-phosphoric acid esters (useful as insecticides, especially against caterpillars), of the general formula $R{\cdot}SO{\cdot}PO(OR_1)(OR_2)$ where R is an alkyl or aryl radical and R_1 and R_2 are alkyl radicals of >4 C-atoms, are produced when an aliphatic or aromatic sulphinic acid is reacted with an *OO*-dialkyl phosphorous acid halide at 40—60° in the presence of an organic acid-binding agent (e.g., pyridine). The prep. of *Me*$_2$ p-*chlorophenylsulphinylphosphonate* is detailed.
E. ENOS-JONES.

Thiocarbamates, bisthio- and bisdithiocarbamates, and compositions containing such substances for treating plants. N.V. Philips' Gloeilampenfabriken (B.P. 811,861, 27.3.56. Neth., 30.3.55).—Compounds $NRR^{I}{\cdot}CS{\cdot}O{\cdot}R^{II}{\cdot}COX$ and $A[NR^{I}{\cdot}CS{\cdot}Z{\cdot}R^{II}{\cdot}COX]_2$ are claimed (R and R^{I} are H, alkyl, or together with N comprise a heterocyclic ring of >6 members; R^{II} is alkylene, aralkylene or arylene; X is NH_2, substituted NH_2, OH or OR^{IV}; R^{IV} is alkyl, aryl, aralkyl or cation; Z is O or S; A is alkylene of 2—3 C). They are effective systemically against fungus diseases of plants. The prep. is detailed of *Na*$_2$ SS′-*di-(carboxymethyl)ethylenebis-dithiocarbamate*.
F. R. BASFORD.

Thiolcarbamates. Stauffer Chemical Co. (B.P. 808,753, 17.1.57. U.S., 17.1.56).—Compounds $NRR'{\cdot}CO{\cdot}SR''$, useful as herbicides (especially pre-emergence herbicides), are obtained by interaction of $NRR'{\cdot}COCl$ with R″SNa (R and R′ are alkyl of <3 C; R″ is alkyl of 1—5 C or alkenyl of 2—5 C). Details are given for the prep. of *Et dipropylthiolcarbamate*, b.p. 135·5—137°/31·5 mm.
F. R. BASFORD.

Dithiophosphoric acid esters. Farbenfabriken Bayer A.-G. (B.P. 811,529, 11.10.56. Ger., 13.10.55).—Compounds $(OR)_2PS{\cdot}S{\cdot}A{\cdot}S{\cdot}B{\cdot}R'$ and insecticidal compositions containing them are claimed (R is alkyl of 1—5 C; R′ is Ph optionally substituted by, e.g., halogen, NO_2 or R; A and B are alkylene of 1—3 C). As an example of the method of prep., that of OO-*diethyl* S-2-*benzylthioethyl thiothionophosphate*, b.p. 171°/0·05 mm., is detailed.
F. R. BASFORD.

Dinitration of o-alkylphenols. Rohm & Haas Co. (B.P. 810,239, 12.9.56. U.S., 16.9.55).—Treatment of a sulphonated alkylphenol with aq. $NaNO_3$ at 70—100° gives good (92—96%) yields of the dinitro deriv. useful as pesticides. One example given is the prep. of 2,4-dinitro-6-hexylphenol.
F. R. BASFORD.

1,1,2-Tri-(1-cyanoethyl)hydrazine. American Cyanamid Co. (B.P. 810,403, 27.2.57. U.S., 26.3.56).—Hydrazine is treated with <3 mol. of $OH{\cdot}CHMe{\cdot}CN$ at 0—100° to give 1,1,2-*tri*-(1-*cyanoethyl*)-*hydrazine*, b.p. 122—123°. The compound is active against nematodes, and compositions are claimed containing it for use in this way.
F. R. BASFORD.

***N*-Morpholinyl-substituted ketones with fungicidal action.** S. A. Farmaceutici Italia (B.P. 810,497, 9.3.56. It., 12.3.55).—Compounds, active against fungi, especially *Aspergillus niger*, *Candida albicans*, *Alternaria solani*, *Penicillium notatum*, *Fusarium dimerum*, *Helmintosporium oryzae*, comprise morpholine hydrochlorides substituted in the 4-position by $[CH_2]_2{\cdot}CO{\cdot}Z{\cdot}OH$ (Z is aromatic or heterocyclic nucleus optionally substituted by alkyl, halogen or NO_2). As an example of prep. is described that of 5-*chloro*-2-*hydroxy*-4-*methyl*-β-*morpholinopropiophenone hydrochloride*, m.p. 194—195°.
F. R. BASFORD.

Fungicides. Farbenfabriken Bayer A.-G. (B.P. 810,044, 27.1.56. Ger., 4.2.55).—A compound $X{\cdot}R{\cdot}NCS$ (X is NH_2, Schiff's base, hydrazone or a salt thereof when R is phenyl, or X is NH_2, Schiff's base, hydrazone or salt thereof, CO_2H or $CONH_2$ when R is naphthyl residue) is compounded with a carrier (viz., solid or water containing emulsifier), to provide a fungicidal composition. In an example, p-$CO_2H{\cdot}C_6H_4{\cdot}NCS$ (0·15) or its Zn salt is admixed with water (100 pt.) and benzylhydroxy diphenyl polyglycol ether to provide a spray capable of protecting greenhouse potato plants against *Phytophthora* infection.
F. R. BASFORD.

Chlorinated aryloxybutyric acids. Monsanto Chemicals Ltd. (Inventor: J. P. Brown) (B.P. 810,154, 28.11.56).—An aryloxybutyric acid is chlorinated at 60—150° (molten state) with Cl_2 or SO_2Cl_2. Thus Cl_2 is passed during 2·5 hr. at 65—80° through (molten) γ-phenoxybutyric acid, then the crude product is cooled and recrystallised from benzene, to give a product, m.p. 104—112°, similar to commercial (2,4-dichlorophenoxy)butyric acid, useful as selective herbicide.
F. R. BASFORD.

Unsaturated polycyclic sulphites. Farbwerke Hoechst A.-G. (B.P. 810,602, 18.4.55. Ger., 17.4. and 18.6.54).—Compounds obtained by the action of $SOCl_2$ on a bicyclo[2,2,1]hept-2-ene or -2,5-diene with $CHR{\cdot}OH$ (R is H or Me) substituent in the 5- and 6-positions, are useful as pesticides (fungicides and insecticides). The prep. is described of the *cyclic sulphite* of *hexachloro*-5,6-*di*-(α-*hydroxyethyl*)*bicyclo*[2,2,1]*heptadiene*, m.p. 93—95°.
J.A.C. ABSTR.

Animal Husbandry

Comparison of the digestibility of forages by cattle and by sheep. R. W. Swift and J. W. Bratzler (*Pa agric. Exp. Sta.*, 1959, Bull. 651, 5 pp.).—The apparent digestibility of the dry matter, protein and energy of 28 lots of forage (lucerne cut at various stages of maturity, orchardgrass and bromegrass at various stages of maturity and grown with varying levels of N, mixed grasses and legume hays) were determined with cattle and sheep at various locations. There were no significant differences between the digestive capacity of the two animals for any of the constituents of any of the forage samples.
A. H. CORNFIELD.

Comparison of nutritive value of lucerne hay with bromegrass and reed canary grass hays at various levels of nitrogen fertilisation. K. M. Barth, C. W. Vander Noot and J. L. Cason (*J. Nutr.*, 1959, **68**, 383—391).—Bromegrass hays (**I**) grown with 25, 125 and 225 lb. of N/acre and reed canary grass hays (**II**) with 0, 100 and 200 lb. of N/acre were compared with lucerne hay (**III**). Increase in N-fertilisation increased the protein from 7·9 to 15·2% in **I** and 12·6 to 20·1% in **II**; at high N-levels in **I** there was also an increase in gross energy and ether extract while fibre and N-free extract decreased. In **II**, no similar trend was observed but a caveat is given relating to variability. In both species a statistically significant increase in apparent digestibility of protein was noted with increased N-fertilisation. The digestible energy coeff. also increased with higher levels of N in **I** but the differences were less marked and not significant at all levels. Fibre digestibility for the grass hays were considerably higher than with **III**. (19 references.)
C. V.

Effects of foliar application of urea on nutritive value of some grass hays. W. G. Merrill (*Dissert. Abstr.*, 1959, **20**, 1508).—Applications of urea to the soil or to the leaves of grass crops increased the crude protein content of the hay, and the apparent digestibility of the crude protein, as determined by digestion trials with sheep and young bulls, by *in vivo* digestion trials with the rumen micro-organisms of a rumen-fistulated steer, and by *in vitro* digestion by rumen micro-organisms. The same effect was found with hay supplemented with urea at time of feeding.
M. D. ANDERSON.

Pangola grass (*Digitaria decumbens*, Stent) in the U.S. Virgin Islands. A. J. Oakes, R. M. Bond and O. Skov (*Trop. Agric.*, 1959, **36**, 130—137).—The agronomic characteristics, diseases and parasites, establishment, response to fertility, carrying capacity and relation to beef production, are reviewed.
E. G. BRICKELL.

Conservation and digestibility of some conserved fodder crops for dry season feeding in Ghana. T. J. Lansbury (*Trop. Agric.*, 1959, **36**, 305—308).—Eight green fodders were examined. Centrosema, green maize and fodder millet provided adequate energy and protein for growth and milk production in cattle. Guatemala grass, Guinea grass and eight-week millet and velvet bean mixture need slight protein supplementation but might be adequate for conditions of extensive production. Mature Bana grass, and ten-week millet and velvet bean mixture definitely need supplementation.
E. G. BRICKELL.

Critical study of energy determination in fresh and dried cow faeces. H. Fenner and J. G. Archibald (*J. Dairy Sci.*, 1959, **42**, 1995—2001).—An accurate method of determining energy in fresh faeces is described. Moisture determination by freeze-drying, toluene distillation and oven drying are compared. The energy contents of freeze-dried, oven-dried and fresh faeces differed by $<2\%$.
S. C. JOLLY.

Silage fermentation. B. J. Walker (*Dissert. Abstr.*, 1959, **20**, 1510—1511).—Freshly chopped grass–lucerne mixtures were ensiled in plastic bags, from which air was removed; samples were analysed at intervals. No propionic acid was formed. Butyric acid was formed in untreated silage, but not when Na metabisulphite or molasses was added. With molasses, there was a vigorous lactic acid fermentation. Bisulphite inhibited acid production. Titratable acidity and/or pH may be as useful as lactic acid for evaluating natural silage fermentations, but neither are adequate for determining the degree of preservation of bisulphite-treated silage.
M. D. ANDERSON.

Storage structures for grass silage. C. C. Zoerb, H. G. Young, H. H. DeLong and D. L. Moe (*S. Dakota agric. Exp. Sta.*, 1959, Bull. 477, 35 pp.).—The performance of upright silos, bunker silos, trench silos, and stacks for ensiling lucerne–bromegrass was studied.
A. H. CORNFIELD.

Composition and digestibility of some conserved fodder crops for dry-season feeding in Ghana. T. J. Lansbury (*Trop. Agric.*, 1959, **36**, 65—69).—Analyses and digestibility coeff. (bullocks) of Guinea corn and millet silages are recorded. Energy values were adequate but protein levels were too low for bullocks. A. G. POLLARD.

Gossypol in cottonseed meals. H. R. Halloran and G. C. Cavanagh (*Poultry Sci.*, 1960, **39**, 18—25).—There was poor correlation between free gossypol and "Available Gossypol Units" (AGU) (*J. agric. Fd Chem.*, 1954, **2**, 982) in 45 cottonseed meal samples, indicating that so-called "degossypolised" meals are not necessarily safe for inclusion in laying rations. Properly processed meals could be produced having less than 0·3% AGU and could be included in layer rations up to 10% of the diet. Eggs from hens receiving such diets did not develop objectionably coloured yolks after 3—6 months' storage at 0·5°, whilst storage at 10° produced a few objectionably coloured yolks. A. H. CORNFIELD.

Relations between rumen acids and fat metabolism of ruminants fed on restricted roughage diets. P. J. Van Soest and N. N. Allen (*J. Dairy Sci.*, 1959, **42**, 1977—1985).—Feeding restricted amounts of roughage with high levels of concentrates significantly reduced milk fat % and blood-ketone levels and increased propionic acid levels in the rumen fluid of lactating cows and goats. Arterial blood-acetic-acid levels and the arterial–mammary difference were decreased by ground and restricted-roughage feeding. Milk fat % was reduced by feeding Na acetate and more so by Na propionate. A hypothetical mechanism of milk-fat depression based on anti-ketogenic properties of propionic acid is discussed. S. C. JOLLY.

Salt requirements of dairy cows. S. E. Smith and P. D. Aines (*Cornell agric. Exp. Sta.*, 1959, Bull. 938, 26 pp.).—Lactating cows on a diet of grass–legume hay, maize silage–cereal mixture and linseed meal were fed 0—120 g. of NaCl per cow per day. Cows receiving no added NaCl or 15 g. of NaCl per day soon showed depraved appetites and decreased urinary excretion of Na and Cl^-. After one year unsupplemented cows began losing body wt. and declined in milk production. Some cows developed other symptoms and died. Affected cows quickly recovered when given NaCl. Cows producing about 11,000 lb. of milk per annum required approx. 30 g. of supplementary NaCl per cow per day. Total Na requirement was 21·3 g. per cow per day. Cows deficient in NaCl produced milk of higher ascorbic acid content than did normal cows. A. H. CORNFIELD.

Preferences of cattle for certain flavours and their effect on palatability of salt mixtures. O. J. Stubbs (*Dissert. Abstr.*, 1959, **20**, 1509—1510).—Calves 8—16 weeks old generally preferred 1 or 2% solutions of sucrose to plain water, were indifferent to dil. aq. saccharin, and disliked 0·05—0·1N-HCl and 0·1% quinine. Intake of salt and mineral mixtures varied from 0·1 to ⩽4 oz. per day; addition of the anthelminthic phenothiazine much reduced intake. Sugar or dehydrated molasses or carob flour were the best materials for masking the unpalatability of phenothiazine. M. D. ANDERSON.

Causes and prevention of reproductive failures in dairy cattle. I. Influence of underfeeding and overfeeding on growth and development of Holstein heifers. A. M. Sorensen, W. Hansel, W. H. Hough, D. T. Armstrong, K. McEntee and R. W. Bratton. **II. Influence of underfeeding and overfeeding from birth to 80 weeks of age on growth, sexual development and semen production of Holstein bulls.** R. W. Bratton, S. D. Musgrave, H. O. Dunn and R. H. Foote (*Cornell agric. Exp. Sta.*, 1959, Bull. 936, 51 pp.; Bull. 940, 45 pp.).—Results are presented and discussed. A. H. CORNFIELD.

Comparison of zero, moderate and liberal levels of grain feeding for lactating cows on permanent, improved and supplemental pasture. D. R. Dowden, D. M. Seath, B. F. Brown and D. R. Jacobson (*J. Dairy Sci.*, 1959, **42**, 1960—1965).—Feeding moderate (5—6 lb.) or liberal (10—12 lb.) amounts of grain daily to lactating cows on different types of pasture had no significant effect on milk production or body wt. Grain feeding reduced consumption of pasture dry matter, but increased that of total dry matter. The average dry matter digestibility of orchard grass–ladino clover pasture, with and without Sudan grass, was greater than that of Kentucky bluegrass–white clover pasture. S. C. JOLLY.

Irrigated Sudan grass and millet for forage and feed. R. W. VanKeuren and W. W. Heinemann (*Wash. agric. Exp. Sta.*, 1959, Bull. 605, 9 pp.).—Within 6 weeks of planting, Sudan grass and pearl millet under irrigation provided abundant forage for beef cattle. Yearling steers grazing these species made wt. gains comparable with those grazing orchard grass or tall fescue. A. H. CORNFIELD.

Production and growth of dairy cows reared on silage or hay rations. J. W. Thomas, J. F. Sykes and L. A. Moore (*J. Dairy Sci.*, 1959, **42**, 1949—1954).—Heifers reared on lucerne silage weighed less at 2 years of age and produced slightly, but not significantly, less milk and butterfat during their entire first lactation than did those reared on hay or hay and silage. At later ages, animals in both groups were similar in wt. S. C. JOLLY.

Intake and value for milk production of oat silages ensiled at three stages of maturity and preserved with sodium metabisulphite. F. A. Martz, C. H. Noller, D. L. Hill and M. W. Carter (*J. Dairy Sci.*, 1959, **42**, 1955—1959).—For optimum intake and value for milk production, oats should be ensiled at the boot stage of maturity. Metabisulphite is a suitable preservative. S. C. JOLLY.

Carbonated vegetable phospholipids as additives to rations for cattle. C. M. Kincaid, W. S. Wilkinson, J. C. Taylor, K. W. King and W. E. C. Moore (*Va agric. Exp. Sta.*, 1959, Tech. Bull. 139, 19 pp.).—Addition of soya-bean, cottonseed or gossypol-detoxified cottonseed (1·5%), or cottonseed phospholipid (0·75—3·0%) to beef cattle dry-lot rations increased wt. gains and feed efficiency by 6—7%. The treatments had no effect on dressing %, carcass quality or plasma levels of fat-sol. vitamins, except possibly to increase vitamin E, probably due to their tocopherol content. The materials had no effect on cellulose digestion, fatty acid production or N metabolism when added to rumen fluid *in vitro*. A. H. CORNFIELD.

Lucerne hay and dehydrated lucerne meal in milo and sorghum silage rations for fattening calves. L. S. Pope, R. MacVicar, L. Walters and V. G. Heller (*Okla. agric. Exp. Sta.*, 1959, Bull. 522, 16 pp.).—A ration containing rolled milo, cottonseed meal, sorghum silage and minerals was adequate for fattening steer calves. Replacing 25—50% (protein basis) of the cottonseed meal with lucerne hay or 25—100% of the cottonseed meal with dehydrated lucerne meal pellets did not improve wt. gains or feed efficiency and had little effect on carcass quality. There were indications of vitamin-A deficiency when the basal ration was low in carotene. A. H. CORNFIELD.

Cane molasses and urea fed with early- and late-cut hay in a ration for dairy heifers. W. G. Merrill, S. Lovejoy, K. L. Turk, K. E. Harshberger, J. K. Loosli and G. W. Trimberger (*Cornell agric. Exp. Sta.*, 1959, Bull. 937, 12 pp.).—Rations in which cane molasses replaced corn-and-cob meal on an equal TDN basis were satisfactory for growing dairy heifers, but intake of hay was increased. Gains with rations containing molasses + urea were lower than those with molasses + either soya-bean oil meal or corn-and-cob meal or with soya-bean oil meal alone, especially when fed with late-cut hay. A. H. CORNFIELD.

Oestrogenic activity in milk of cows and the bile of calves fed low levels of stilboestrol. E. H. Herrick, C. Paulson, R. Baron and C. B. Browning (*J. Dairy Sci.*, 1959, **42**, 1966—1969).—No oestrogenic activity was detected in milk from cows fed 10 mg. of stilboestrol per 1000 lb. of body wt. daily; activity equiv. to 46 μg. of stilboestrol was excreted daily in the bile of calves fed similarly. No activity was detected in the milk at any stage of the oestrus cycle in untreated cows or in the bile of untreated calves. S. C. JOLLY.

Partition of riboflavin in cow's milk. V. V. Modi, E. C. Owen and R. A. Darroch (*J. Dairy Res.*, 1959, **26**, 277—280).—As lactation progressed, the total riboflavin content of milk from six Ayrshire cows declined gradually from initial values of 1·09 to 0·86 μg. per ml. at rates of 0·01 to 0·003 μg. per ml. daily. Treatment with thyroxine stimulated milk yield and heart rate, and prevented the rise in phosphatase concn. in the milk as lactation progressed, but it did not affect either the concn. or the partition of riboflavin between the protein-bound and the free forms. Reasons are suggested for the marked contrast between the effect of thyroxine in increasing the phosphorylation of thiamine and the absence of any such effect on riboflavin. (15 references.) S. C. JOLLY.

Energy value of various feedstuffs for young pig. B. G. Diggs (*Dissert. Abstr.*, 1959, **20**, 1505—1506).—Fraps and Carlyle's algebraic method for determining the maintenance requirement of young pigs and the productive energy values of foodstuffs gave variable results; using a constant figure for maintenance gave more consistent productive energy values. In balance experiments with pigs weighing about 34 lb., digestible energy (**I**), metabolisable energy (**II**), and **II** adjusted for N balance (**III**), were determined for 18 foodstuffs; **III** was calculated as 6·77 kg.-cal. per g. of urinary N, for diets containing 35% or more of protein. The sp. gr. of a live pig is not usefully correlated with kg.-cal. per g., but regression equations were obtained for predicting kg.-cal. per g. from fat or water content of the carcass. M. D. ANDERSON.

Prevention of anaemia in baby pigs. W. G. Pond (*Okla. agric. Exp. Sta.*, 1959, Bull. 939, 14 pp.).—Normal haemoglobin, hematocrit, and growth of suckling pigs were obtained by injection of an Fe–dextran complex (0·1 g. of Fe) at 3 and 10 days of age. Daily spraying of the sow's udder with $FeSO_4$ solution (to give 10 g. Fe

per sow daily) was also satisfactory, but more laborious. Oral Fe–Cu–Co tablets at 3 and 10 days of age (giving 0·6 g. of Fe) did not support blood values so well, but maintained normal growth. A. H. CORNFIELD.

Treatment of new-born pigs with iron dextran. W. E. Crawley and H. J. MacDiarmid (*N.Z. J. agric. Res.*, 1959, **2,** 1121—1122).—Administration of Fe-dextran to piglets prevented the marked fall in haemoglobin suffered by controls but did not affect live-wt. gains. Where there is access to pasture and soil, the use of Fe-dextran for routine treatment of new-born piglets is not recommended. K. R. BUTLIN.

Comparison of dried skim milk and white fish meal as protein supplements for fattening pigs. IV. Further studies with pigs fed unrestricted amounts of whey under commercial conditions. R. Braude, K. G. Mitchell, A. S. Cray, A. Franke and P. H. Sedgwick (*J. Dairy Res.*, 1959, **26,** 238—247).—The performances of fattening pigs were not significantly different when fed unrestricted amounts of whey supplemented daily with 3 lb. (reduced to 2 lb. at 3 weeks of age) of a basal meal containing either 10, 7 or 5% of white fish meal or 10% of dried skim milk. Either 10% of the fish meal or 15% of the milk powder should be the minimum amounts of these protein sources in a basal meal fed with unrestricted amounts of whey, and ≮2 lb. of the meal should be given daily. Possible effects of season on whey consumption are discussed in relation to the effects of whey acidity on performance of the pigs. S. C. JOLLY.

Effects of feeding wheys of different ages to fattening pigs. A. C. Dunkin (*N.Z. J. agric. Res.*, **2,** 1111—1120).—Pigs fed whey obtained daily showed somewhat, but not markedly, better growth rates and food conversion efficiencies than when the whey was supplied on only 4 days each week. Addition of bleaching powder to reduce development of acid had no beneficial effect. K. R. BUTLIN.

Carbohydrate metabolism of sheep. IX. Metabolic effects of glucose and glycerol in undernourished pregnant ewes and in ewes with pregnancy toxaemia. R. L. Reid (*Aust. J. agric. Res.*, 1960, **11,** 42—57; cf. *ibid.*, 1959, **10,** 81).—Experimental data recorded support the view that pregnancy toxaemia is associated with depressed glucose metabolism. A. G. POLLARD.

Metabolic aspects of urea and carbohydrates versus plant protein for sheep. D. Drori (*Dissert. Abstr.*, 1959, **20,** 1506).—Sheep were fed a basal diet of chopped hay, molasses and minerals, with 70% of the N supplied by supplements of glucose + urea + Na_2SO_4 (i), maize starch + urea + Na_2SO_4 (ii), or soya-bean oil-meal and maize (iii). With equicaloric diets, N retention was highest on diet (iii). Blood urea and non-protein-N increased on (i) and (ii), and urea was detected in 22 of 56 samples of rumen liquid. Blood-acetone and rumen volatile fatty acids were highest on (iii). Consistent increases in blood-glucose after feeding were found only on (iii). Diets (i) and (ii) were consumed more slowly than (iii). M. D. ANDERSON.

Plasma non-esterified fatty acids in sheep. E. F. Annison (*Aust. J. agric. Res.*, 1960, **11,** 58—64).—The importance of these plasma-acids in sheep metabolism is emphasised by data recorded on fasting pregnant and non-pregnant sheep and on the effects of injected glucose and insulin. A. G. POLLARD.

Effects of thyroxine on live weight, metabolism and wool growth of Romney ewes. A. H. Kirton, E. Cresswell, F. R. Cockrem and G. W. Butler (*N.Z. J. agric. Res.*, 1959, **2,** 1143—1158).—Injection of thyroxine (5 and 1 mg./day) increased O_2-consumption, heart and respiration rates of 12 Romney ewes. Live wt. were reduced with 5 mg./day but unaltered by 1 mg./day; rectal temp. remained normal; 0·75 mg./day increased heart rate but not live wt.; 0·5 mg. affected neither. Thyroxine injections markedly increased wool growth. Thyroxine implantations produced no effects. (24 references.) K. R. BUTLIN.

Effect of shearing time on wool. IV. Effect on fleeces. V. Effect on processing. L. F. Story and (IV) D. A. Ross (*N.Z. J. agric. Res.*, 1959, **2,** 1096—1103, 1104—1110).—IV. Tests on flocks of Romney ewes (4 successive years) and Corriedales (2 years) show that pre-lambing shorn wool is much superior to post-lambing shorn wool in strength, soundness and freedom from tenderness and cotting. No evidence was found that pre-lambing fleeces are lighter or shorter than post-lambing fleeces.

V. In processing tests, wools shorn before lambing gave a higher yield, less noil, a longer top and stronger yarn than post-lambing wool shorn from the same flock. Pre-lambing wool is not inferior in processing performance. K. R. BUTLIN.

Storage of dieldrin in tissues [A] and its excretion in milk of dairy cows fed dieldrin in their diets: [B] of steers, hogs, lambs and poultry fed dieldrin in their diets. [C] Insecticide residues in milk of dairy cows fed insecticides in their daily ration. N. Gannon, R. P. Link and G. C. Decker (*J. agric. Fd Chem.*, 1959, **7,** 824—826, 826—828, 829—832).—[A] Dieldrin (**I**) fed to cows at levels of 0·1, 0·25, 0·75 and 2·25 p.p.m. in the feed appeared in the milk after 6—12 weeks of continuous administration at average levels of 0·02, 0·06, 0·11 and 0·28 p.p.m. respectively; in some cows **I** appeared in the milk within 2 weeks at the lowest level of intake and within 3 days at the highest level. At the end of the experimental period, **I** was present in renal and body fat at concn. varying from 0·24 p.p.m. at the lowest level of intake to 5·48 p.p.m. at the highest level; the amount of **I** in other tissues was proportional to their fat content.

[B] Dieldrin (**I**) was detectable in the fat of all animals receiving it at levels of 0·1—2·25 p.p.m. in the feed; when detectable in other tissues, the amount was proportional to the fat content of the tissue. For each species, the amount of **I** stored was apparently proportional to the intake. Laying hens stored the greatest amounts, but their eggs contained very little **I.** Levels in the tissues of steers were higher than in those of hogs, which in turn were higher than in those of lambs.

[C] Levels of aldrin (excreted as dieldrin), dieldrin, heptachlor (excreted as its epoxide), Dicophane (DDT) and methoxychlor excreted in the milk and stored in the fat by cows fed these insecticides at various rates in their feed daily for 16 weeks are reported. Aldrin was excreted at approx. twice the rate of dieldrin, 10 times that of Dicophane, 20 times that of heptachlor and 1600 times that of methoxychlor. Propensity for storage in fat followed the same order with variation in magnitude. Wt. gains and milk production were unaffected by the treatments, and no pathological lesions occurred in tissues or organs. S. C. JOLLY.

Heptachlor residues on maize stover in relation to dairy cattle feeding. R. E. Johnsen, P. A. Dahm, H. W. Rusk, M. L. Fairchild and A. E. Freeman (*J. econ. Ent.*, 1960, **53,** 19—22).—The butterfat from the milk of cows fed on maize stover, 133 days after application of 5% granular heptachlor, contained no heptachlor epoxide when analysed spectrographically. (12 references.) C. M. HARDWICK.

Metabolism and residues of ^{32}P-labelled Delnav in a Hereford steer. F. W. Plapp, jun., W. S. Bigley and D. I. Darrow (*J. econ. Ent.*, 1960, **53,** 60—64).—Most of the spray remained on the hair and was absorbed only slowly. Paper chromatography separated diethyl-phosphoric, diethyl phosphorothioic, and diethyl phosphorodithioic acids as the main metabolites. No residues were found in meat and only small amounts accumulated in the fat. The peak radio-activity in the urine was 12 hr. and in the faeces 24 hr. after spraying. The method of application did not affect the metabolism in mice. C. M. HARDWICK.

Influence of dietary protein level and amino-acid balance on pigmentation in the feathers of chicks. G. J. Klain, D. C. Hill, J. A. Gray and E. M. Olsen (*Poultry Sci.*, 1960, **39,** 25—29).—Trials with Barred Plymouth Rock cockerels showed that feather achromatosis was not caused by a low intake of lysine *per se* but occurred when lysine was low and other amino-acids were given in excess. Achromatosis did not occur when a well-balanced diet providing adequate protein for normal growth was supplemented with a large excess of zein or soya-bean protein. A. H. CORNFIELD.

Growth response of chicks to fluid egg yolk. D. E. Greene, H. M. Scott and H. W. Norton (*Poultry Sci.*, 1960, **39,** 7—12).—Addition of 20% fluid egg yolk or 6·5% egg oil to two types of basal diet improved chick growth rate. The responses were less with the basal diet which by itself supported the greater wt. gain. With the less efficient basal diet there was a greater response to egg yolk than to egg oil, this difference probably being due to the protein contributed by the yolk. The growth-promoting effect of egg yolk is probably due to an improvement in the balance of known nutrients rather than to the presence of some unidentified growth factor. A. H. CORNFIELD.

Effect of dietary protein and fat on changes of cholesterol level in chicks. M. G. Kokatnur (*Dissert. Abstr.*, 1959, **20,** 1733).—Chicks were fed on different purified complete diets. After 4 weeks, serum- and carcass-cholesterol were inversely related to protein intake. At low protein levels, serum-cholesterol was not affected by the fat content of the diet, but at higher protein levels, values increased with the fat content of the diet. Carcass-cholesterol increased with the fat content of the diet if the fat was "hard" (e.g., lard), but decreased if it was "soft" (e.g., maize oil). Serum- and carcass-cholesterol and carcass-fat decreased as the energy/protein ratio of the diet fell. The effects of type of fat on serum-cholesterol were insignificant at a low energy/protein ratio, and pronounced at a high energy/protein ratio. M. D. ANDERSON.

Mineral requirement of the chick with special reference to zinc and magnesium. M. W. A. Moeller, jun. (*Dissert. Abstr.*, 1959, **20,**

1508—1509).—Protein in purified diets for chicks was supplied as soya-bean protein (**I**), dried egg white (**II**) or casein (**III**), with amino-acid supplements. For optimum growth and feed conversion, chicks on the 3 diets required respectively 27—31, 10—12 and <14 p.p.m. of Zn. Zn in soya-bean protein is not fully available, and the protein accentuates the need for Zn. On Zn-deficient diets, the concn. of Zn in the skeleton but not that in growing feathers paralleled the Zn content of the diet. A chick bioassay showed that the availability of Zn in maize and soya-bean meal is less than and that in distillers' dried solubles is equal to that in $ZnCO_3$. On diets **I** and **III**, the Mg requirements of the chick for max. growth were respectively 400—465 and 390—470 p.p.m.
M. D. ANDERSON.

Changes with age in tissue levels of sodium and potassium in the fowl. E. M. Kravis and M. R. Kare (*Poultry Sci.*, 1960, **39,** 13—15).—The Na and K levels in the brain, skin, muscle, liver, plasma and whole blood of White Leghorns fluctuated somewhat from hatching to about 12—18 days of age, but were fairly constant thereafter to 35 days of age. In particular muscle-K increased considerably, whilst muscle-Na decreased considerably, from hatching to 12 days of age. A. H. CORNFIELD.

Carotene content in the yolk of eggs of Green-leg, Sussex and Leghorn hens. A. Kolataj (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E,** 517—532).—No essential differences in the carotene group were found between the Leghorn and Sussex breeds and in the three breeds there was no correlation between the amount of pigments in the egg yolk and the rate of egg-laying. In Green-leg and Leghorn breeds the correlation values between xanthophyll and carotene groups were negative ; no such dependence was observed in the Sussex breed. The greatest amounts of pigments were found in the Sussex eggs although darker yolks were produced by Green-legs. In the breeds studied genetic differences exist in the uptake and production of carotenoid pigments. (33 references.) (From English summary.) E. G. BRICKELL.

Influence of carotenoid pigments and vitamin A on hatchability of eggs and growth of chickens hatched from them. A. Kolataj (*Ann. Univ. M. Curie-Sklodowska*, 1957, **12E,** 489—516).—Fowls should be allowed to feed on the range and to be given green crops in their diet. Addition of cod-liver oil to the daily ration is also desirable. The content of vitamin A in the egg yolk influences the % of embryos dying in shell during incubation. The highest growth rate was observed in chickens hatched from eggs laid by range-fed hens ; the content of vitamin A in the egg yolk influences the % of fertilisation. (55 references.) (From English summary.)
E. G. BRICKELL.

Bacteriological and serological studies of *Escherichia coli* serotypes with calf scours. P. J. Glantz, H. W. Dunne, C. E. Heist and J. F. Hokanson (*Pa agric. Exp. Sta.*, 1959, Bull. 645, 22 pp.).—A study was made to determine whether certain serotypes of *Esch. coli* were of etiological significance in calf scours. The effects of agglutinins, the bactericidal activity of calf and dam serum and the significance of colostrum in experimental infection of calves with *Esch. coli* were also investigated. A. H. CORNFIELD.

Cattle ticks, *Boophilus microplus*, resistant to DDT, BHC and dieldrin. B. F. Stone and L. G. Webber (*Aust. J. agric. Res.*, 1960, **11,** 105—119).—In an area in which ticks had developed resistance to DDT, control by γ-BHC or dieldrin (0·5% w/v, in each case) became less effective within a year. The efficiency of diazinon did not diminish in 1 year. A. G. POLLARD.

Organophosphate systemics as sprays and feed additives for cattle grub control. E. S. Raun and J. B. Herrick (*J. econ. Ent.*, 1960, **53,** 125—126).—From 76—98% control of *Hypoderma* spp. was obtained by oral administration of Ronnel (15 mg./kg. for 7 days or 25 mg./kg. for 6 days). Bayer 21/199 as a suspension or emulsion gave similar results. No differences in wt. gain were found.
C. M. HARDWICK.

Systemic insecticides for the control of *Gasterophilus* bots in horses. R. O. Drummond, J. B. Jackson, E. E. Glass and B. Moore (*Agric. Chemic.*, 1959, **14,** No. 12, 41—43, 100).—Addition of Dipterex (*OO*-dimethyl 2,2,2-trichloro-1-hydroxyethylphosphonate, 0·04 g. per kg. of diet) to the feed of horses gave good control of two species of bots. Some of the other materials tested were toxic to horses at doses required for effective control of bots.
A. H. CORNFIELD.

Control of *Myocoptes musculinus* on guinea pigs. H. G. Sengbusch (*J. econ. Ent.*, 1960, **53,** 168).—Dipping 3 times in 0·2% DMC or 2% Aramite controlled mange mites. After disinfestation of the cages the animals remained free for 8 months. C. M. HARDWICK.

Selective toxicity and animal systemic effectiveness of several organophosphates. U. E. Brady, jun., H. W. Dorough and B. W. Arthur (*J. econ. Ent.*, 1960, **53,** 6—8).—The toxicity of 17 compounds administered orally and subcutaneously to rats, orally to rabbits and topically to houseflies is listed. Of the compounds six were effective against *Cimex lectularis* and *Amblyomma maculatum* feeding on treated rabbits. Results are related to structure. (11 references.) C. M. HARDWICK.

Streptimidone, a new antibiotic. D. L. Kohberger, M. W. Fisher, M. M. Galbraith, A. B. Hiltegas, P. E. Thompson and J. E. Ehrlich (*Antibiotics & Chemotherapy*, 1960, **10,** 9—16).—*In vitro*, streptimidone and dihydro-deriv. were as effective as emetine against *Entamoeba histolytica* ; *in vivo*, 5 mg./kg./day cured severe amoebic dysentery in dogs. C. V.

Control of poultry lice and mites with several organic insecticides. R. A. Hoffman (*J. econ. Ent.*, 1960, **53,** 160—162).—Sprays of Co-ral and malathion eliminated three spp. of lice on hens in 5—7 days. Sevin and Delnav took a little longer. All treatments were less effective against mites. Sevin and Dicapthon dusts applied to litter were effective against lice at a lower rate than were malathion or Ronnel. C. M. HARDWICK.

Laboratory and field tests against mites and lice attacking poultry. W. S. Bigley, A. R. Roth and G. W. Eddy (*J. econ. Ent.*, 1960, **53,** 12—14).—In laboratory tests with impregnated cloth, Ronnel was more toxic than Barthrin or malathion to *Ornithonyssus sylviarum* but all three were less effective against *Dermanyssus gallinae*. Dicapthon and Ronnel were more effective than malathion when sprayed on to individual hens. Both Ronnel and malathion gave excellent mite control in field tests on turkeys. Both compounds also controlled *Menacanthus stramineus* on chickens as also did 3,4-dimethylbenzyl chrysanthemumate. C. M. HARDWICK.

Control of the Northern fowl mite, *Ornithonyssus sylviarum*, (C. & F.), with Ronnel, Bayer L13/59 and Bayer 21/199. F. W. Knapp and G. F. Krause (*J. econ. Ent.*, 1960, **53,** 4—5).—All three org. P dusts applied to fowls gave satisfactory control for 28 days. The taste of the eggs was unaffected and no residues were found.
C. M. HARDWICK.

Animal foodstuffs. Farbenfabriken Bayer A.-G. (B.P. 808,717, 4.2.57. Ger., 4.2.56).—There is claimed as a growth-promoting supplement for incorporation (in 0·0005—0·005 wt.-%) into livestock feed a heteroauxin of specific activity on the multiplication and growth of cell of plants, e.g., α-naphthylacetic acid, 4-chloro-2-methylphenoxyacetic acid, 2,4-dichlorophenoxyacetic acid, phenoxyacetic acid, phenylacetic acid or indol-2-ylacetic acid (or salts thereof). F. R. BASFORD.

Protein meal of marine origin. A. J. S. Marstrand (B.P. 810,537, 5.11.56. Norw., 5.11.55).—Protein meal of marine animal origin is conditioned (stabilised) prior to storage by storing in a bin and passing a stream of moist gas (air) at 35—60° through the meal from the bottom to the top. F. R. BASFORD.

Veterinary composition. Imperial Chemical Industries of Australia & New Zealand Ltd. (B.P. 811,974, 20.5.57. Aust., 30.5.56).—A veterinary composition for use in the treatment of footrot in sheep comprises a solution (100) of a quaternary ammonium cationic detergent and antiseptic (10—30 pt.) in an alcohol, e.g., MeOH or EtOH. The active agent is preferably hexadecyltrimethylammonium bromide (in the form of cetrimide B.P.).
F. R. BASFORD.

2.—FOODS

Carbohydrate Materials

Cereals, flours, starches, baking

Factors affecting the consistency of canned cream style sweet maize. D. R. Davis (*Dissert. Abstr.*, 1959, **20,** 2221).—Three varieties of maize were tested in order to determine the effect on their consistency of maturity, the amount of water added, the amount and type of starch added, and the time and temp. of storage. Each variety was shown to have a distinct consistency pattern. There was a direct relationship between the maturity of the maize and the amount of water required. The amount of starch affected the consistency much more than the type of starch. The effect of storage temp. varied with the different varieties of maize.
A. M. SPRATT.

Relative effects of enzymic and physical changes during storage on culinary properties of rice. H. S. R. Desikachar and V. Subrahmanyan (*Cereal Chem.*, 1960, **37,** 1—87).—Samples of freshly milled rice and of rice stored for one year were subjected to chemical, physical and cooking tests and the results compared. The α- and

β-amylases were destroyed during the first 5 min. of cooking. Physical changes during storage result in hardening of structure and improvement in quality of the grain. (11 references.)
J. V. Russo.

Treatment of wheat with ionising radiation. I. Effects on milling quality of hard winter wheat. II. Effects on pentosans and on baking quality of hard winter wheat. W. Seibel and (II) F. K. Finney and M. Milner (*Getreide u. Mehl*, 1960, **10**, 13—16; 16—20).—I. A mixture of hard winter wheats was submitted to γ-irradiation of 0 to 3×10^6 rep. No changes in flour yield, ash content and character, or analytical data of the by-products were found in relation to radiation intensity. The colour of the flour was adversely affected by the γ-irradiation; this can be assumed to be due to a browning reaction. Power consumption in milling diminished with increasing dosage, possibly because of the opening-up of chain linkages in the starch and protein of the endosperm. No differences in degree of fineness of the flour could be found. In view of the growing importance of flour colour in judging milling quality, the changes due to γ-irradiation must be regarded as adverse. (16 references.)

II. With hard winter wheat submitted to γ-irradiation of 0 to 3×10^6 rep both flour and bran, after milling, showed increasing water-solubility of the pentosans, and above $0{\cdot}5 \times 10^6$ rep the water-sol. pentosans lost their gel-forming ability in presence of oxidising agents. In consequence of increased formation of reducing sugar the gas-producing ability of the flour increased with rising dosage, but gas retention in the dough diminished from $0{\cdot}1 \times 10^6$ rep. Weakening of the dough was confirmed by mixogram curves. In baking tests, water absorption, kneading time, loaf vol. and crumb value were progressively diminished, and colour and flavour deteriorated, with rising dosage, although optimum addition of bromate helped to retard loaf vol. change. (33 references.)
C. L. Hinton.

Distribution of minerals and proteins in whole wheat, flour and flour fractions, as influenced by variety, season and fertiliser applications. M. M. S. A. El-Gindy (*Dissert. Abstr.*, 1959, **20**, 1941—1944).—Analyses were made of three varieties of wheat grain grown with various fertilisers, and of gluten, starch and water-sol. fractions of the grain. Yield of grain was increased by N, P and K fertilisers, singly or together, but milling quality was not affected. Mixogram readings varied with protein content. N fertiliser increased gluten content, slightly decreased starch content, and usually decreased ash. P and K together usually increased starch content. P increased ash slightly in 15 of 24 instances. Ash content was highest in the water-sol. fraction, and least in the starch, P was highest in starch and least in water-sol., Mg and K highest in water-sol. and least in gluten. B, Mn, Pb, Al, Mo, Sn, Cu, Ag and Zn were determined in the whole wheat and the fractions. Mo was not detectable in the water-sol. Heavy metals were highest in the gluten; Ca also was highest in gluten. M. D. Anderson.

Fineness measurement of ground flours and flour fractions. The fineness-of-grind gauge. D. J. Stevens (*Milling*, 1960, March 18, Repr.).—A method for measuring the degree of granularity of flour in the sub-sieve range (<0·060 mm.) by the Hegman Gauge is described and illustrated. The sample (1 g.) on a black glazed tile is mixed with paraffin, added dropwise, until the paste falls from the palette knife in a syrupy thread. A little of this paste is placed in the deep end of the channel in the fineness-of-grind gauge. A scraper blade at an angle of 45° is drawn slowly along the gauge. If the sample contains no particles >0·1 mm. the surface of the paste at the deep end is smooth and towards the shallow end scratch marks caused by the coarsest particles are visible. Airborne dust should be excluded. Results are in good agreement with microscopical measurements. E. M. J.

Sedimentation method for determining flour particle size. V. J. Evans (*Cereal Sci.*, 1960, **5**, 40—43).—A method for the determination of flour particle size, based on the rate of sedimentation in methanol and its application to cake-, bread- and hard wheat flours is described. J. V. Russo.

Irradiation of flours from thirteen varieties of wheat. C. C. Lee (*Cereal Chem.*, 1960, **37**, 78—80; cf. J.S.F.A. Abstr., 1959, ii, 39).—The effect of irradiating 13 varieties of wheat, with widely different breadmaking quality, with 700,000 r (γ-rays from ^{60}Co) on gluten recovery, protein content and maltose values is described.
J. V. Russo.

The effect, on the solubility of gluten proteins, of the treatment of flours by γ-rays. A. R. Deschreider (*Fermentatio*, 1959, 347—351).—The solubility of flour proteins in water, aq. K_2SO_4 and alcohol of various strengths is affected in various ways by treatment by γ-rays, viz.: (*a*) in alcohol, diminished; (*b*) in aq. K_2SO_4 increased; (*c*) increased in soluble-in-water proteins of indigenous flours but decreased in those of non-Belgian flours. J. V. Russo.

Vitamin contents of air-classified high- and low-protein flour fractions. C. R. Jones, J. R. Fraser and T. Moran (*Cereal Chem.*, 1960, **37**, 9—18).—The effects of grinding soft and hard English wheat flours on the thiamine, niacin, pyridoxine and pantothenic acid contents of fractions separated by air classification are reported. Various theories for these results are discussed. Thiamine contents of hard flour, fine fractions, are much higher than those of the initial flour. Levels of niacin in the fine fractions are much higher with the hard than soft flours. With the hard flours the major part of the increased contents of pantothenic acid and pyridoxine in the fine fractions is attributable, as with riboflavin, to their increased contents of scutellum and aleurone. (16 references.)
J. V. Russo.

Production of amylose and amylopectin in maize endosperms and potato tubers. S. R. Erlander (*Cereal Chem.*, 1960, **37**, 81—93).—Experiments aimed at explaining the mechanism employed by plants in synthesising starch are discussed. The effects of light and dark and of water evaporation are particularly studied. (21 references.)
J. V. Russo.

Special flours for baking. W. H. G. Wiebols (*Bakkerswereld*, 1959/60, **20**, Repr., 8 pp.).—The American Turbo milling process is described; comparative baking tests have been made with additions of these products to Dutch flours. Additions of 20% of Beevo high-protein (20%) flour increase dough-expansion by 20—30%, and also improve the quality of the bread in other respects. In cake-making tests, the results obtained (especially as regards dough-expansion) are for the American hard wheat cake flour, somewhat better, and for low-protein soft wheat cake flour considerably better than those obtained with Dutch patent flour. P. S. Arup.

Factors contributing to the stability of fat in chilled doughs. C. E. Weir, A. D. Slover, J. D. Parsons and L. R. Dugan (*Food Res.*, 1960, **25**, 120—126).—Pastry doughs prepared with (i) prime lard, (ii) similar lards containing singly and in combination, butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate and citric acid, were stored raw or baked, at −10, 0 or 20°F for 36 weeks. The stability of the fat system was not affected by baking prior to storage at low temp. Stability of the fat was extended by addition of 0·01% of BHA or BHT; or both to a greater extent. All samples showed increased stability during the early part of the storage period and again at 20 weeks except those containing BHA. These showed a tendency to decrease in stability at 12 weeks and increase at 20 weeks. E. M. J.

Effects of additions of fat on gas-retention and baking quality of doughs. A. Schulz (*Brot u. Gebäck*, 1960, **14**, 38—39).—Addition of fat was found to have no significant effect on gas formation in dough, as measured by power requirement. Hardened whale oil, palm oil and premier jus, added to an extent of 0·25—1·0%, improved gas retention and baking qualities. In comparisons at 1% fat level bakery margarine was somewhat less efficacious. The improvement in baking quality due to 1% margarine in a dough prepared from flour giving poor gas retention was considerably increased during standing of the dough up to 2 hr.
C. L. Hinton.

Soya flour as white bread ingredient. I. Preparation of raw and heat-treated soya flours and their effects on dough and bread. II. Fractionation of raw soya flour and effects of fractions in bread. J. M. Pollock and W. F. Geddes (*Cereal Chem.*, 1960, **37**, 19—29, 30—54).—I. The analysis of commercial and experimental soya flours and the effect of their addition to doughs on farinograph curves (**I**) and on bread baking tests are described. Inclusion of raw soya flour (1—5%) gave **I** the characteristics of a stronger flour. In baking tests using $KBrO_3$ (1 mg./100 g. of flour), 1% of raw soya flour improved the bread, but higher levels decreased loaf vol. (16 references.)

II. A method of fractionation of unheated defatted soya flour by solvent separation, dialysis and pptn. is described. The analysis of these fractions and the effect of their addition to bread doughs containing various levels of bromate improvers are discussed. The beneficial effect of 1% raw soya flour observed in baking tests at a low bromate level, and destroyed by heat treatment may be caused by the presence of the heat-labile fraction (XVIII). Baking tests on fractions obtained revealed widely different effects of heat. At the higher bromate levels used, raw, extracted soya flour itself was improved by heat. The baking quality of soya flour depends on the baking quality of individual fractions and differential effects of heat and bromate level. (27 references.) J. V. Russo.

Volatile aromatic compounds in fresh bread. L. Wiseblatt and F. E. Kohn (*Cereal Chem.*, 1960, **37**, 55—66).—The recovery of volatile compounds from freshly baked bread by high-vac. distillation is described. The carbonyl compounds thus produced were converted into their 2,4-dinitrophenylhydrazones, separated by adsorption chromatography on silicic acid, and identified. Volatile

acids, ethanol, furfural, croton aldehyde and $\alpha\beta$ dicarbonyls were also estimated. The addition of these distillates or of a synthetic blend of the compounds to a chemically leavened bread was not found to enhance the flavour. (21 references.) J. V. Russo.

Intercomparison of farinograph absorption obtained with different instruments and bowls. I. Hlynka (*Cereal Chem.*, 1960, **37**, 67—70).—Experiments aiming at obtaining comparable absorption mobility data with the same flour for two different farinographs and three different mixing bowls are described. J. V. Russo.

Crumb-colour of bread. A. W. Croes (*Bakkerswereld*, 1959/60, **20**, Repr., 7 pp.).—Measurements of reflecting capacity are made by means of a Photovolt reflectometer. A circular slice of bread, surrounded by a white-enamelled standard reflecting surface is supported on a rotating glass plate situated above the photoelectric apparatus. A device is included for shifting either the bread or the standard surface into the measuring position. The combined result of three measurements with light through a tristimulus filter is calculated (for each surface) by a previously published formula (cf. Croes, *Chem. Weekbl.*, 1959, **55**, 12). Max. points are given for a "pleasant" white, corresponding with a range between chalky and creamy white. P. S. Arup.

Measurement of bread-crumb colour. A. W. Croes (*Brot u. Gebäck*, 1960, **14**, 21—27).—An apparatus for objective measurement of the "whiteness" of bread crumb is described. Standard slices cut from the crumb are revolved eccentrically over the measuring head of a photo-electric reflection apparatus. Successive readings are taken with three filters in position, green, amber and blue, and the readings are then related to standard MgO brightness *via* a calibration ring of known MgO value. Whiteness (W) is calculated from the formula $W = G - A + B$, where G, A and B are the readings (as % MgO) with the three filters. Results for breads baked from series of mixed flours show that a whiteness measurement must take into account not only the "brightness", given by a reflection measurement with the green filter, but also the yellowness, given by the amber and blue filters. C. L. Hinton.

Effect of temperature on stability of frozen cakes. J. W. Pence and M. Heid (*Food Technol.*, 1960, **14**, 80—83).—The period required for detectable organoleptic changes to occur in each of five kinds of commercial cakes held at different refrigerated or sub-freezing temp. was studied. Texture changes were generally detected before changes in flavour. Yellow and chocolate layer cakes showed good stability at 10°F and better at 0°F but below 0°F these and layer cakes deteriorated. Layer cakes were the most stable of the types studied. All types showed highly significant differences from freshly baked cakes within four weeks at 0°F. Their quality was still quite good; they were superior to day-old unfrozen cakes. (16 references.) E. M. J.

Bakery products. Eastman Kodak Co., Assee of N. H. Kuhrt and M. I. Vangraafeiland (B.P. 810,525, 1.11.56. U.S., 3.11.55).—A bakery composition, which on baking affords a product (cake, etc.) of improved vol., texture and grain, comprises a baking mix (bread dough or cake batter) and 0·1—3 wt.-% of a monoglyceride composition consisting of higher triglyceride having fat-forming fatty acid radicals (50—95) and purified, concentrated monopalmitin and/or monostearin (5—50 wt.-%) such that >25 wt.-% of the monopalmitin and/or monostearin is present in the form of uniformly dispersed needle-like microcrystals. F. R. Basford.

Sugars and confectionery

Experimental study of diffusion [of beet sugar]. P. Devillers and M. Loilier (*Industr. aliment. agric.*, 1959, **76**, 851—866).—Rate of diffusion, sugar losses and purity of juice, influence of temp., time, size of cossettes, character of beets and addition of various chemicals were studied. After an initial phase, a rate of diffusion logarithmically constant for a given temp. was attained. The influence of other factors on the rate was measured in terms of this constant rate expressed as a half-diffusion time. Temp. increased the rate regularly above a critical point between 60 and 70°; below this, a kind of pre-diffusion occurred increasing rapidly between 50 and 60°. Rate of diffusion was inversely proportional to the cross-section for cossettes of constant length. Added chemicals in practicable amounts had little effect on rate and purity. Results under standard conditions (3 hr. at 70°) were very variable for beets harvested on the same date, as regards rate of diffusion, purity and sugar losses. A study of conditions affecting the quality of beets is suggested. C. L. Hinton.

Solubility of calcium oxalate in [beet] sugar juice. M. Roche (*Industr. aliment. agric.*, 1959, **76**, 949—953).—The formation of Ca oxalate (**I**) during purification of beet sugar juice and its deposition on tubes is discussed. The solubility of **I** is increased in presence of excess $CaCO_3$, this excess is removed by carbonatation, but the pptn. of **I** is very slow even in the hot juice in presence of K^+. Increase in sucrose concn. tends to diminish solubility of **I** and this is not increased in presence of 0·02% KCl. At this concn. of K^+ relatively slow pptn. continues even after the last evaporation. Na tripolyphosphate has been used in min. quantity (4 mg./l.) sufficient to prevent deposition of **I** incrustation. E. M. J.

Separation of sugars on ion exchange resins. J. K. N. Jones, R. A. Wall and A. O. Pittet (*Chem. & Ind.*, 1959, 1196).—Polyhydric org. mol. are adsorbed on ion-exchange resins; this suggested that mixtures of sugars could be separated by columns of these materials. Columns (100 × 2 cm.) of Dowex 50W separated a mixture of sucrose, raffinose and glucose (200 mg. each) into chromatographically pure components. With various carbohydrate mixtures, elution proceeded in descending order of mol. size with free sugars; with methylated sugars the reverse is found, the most highly methylated fraction being the last to be removed from the column. Hydrolysates of methylated cherry gum and methylated starch were separated into fractions according to the degree of methylation 2,3,5-tri-*O*-methyl-L-arabinose and 2,3 : 4,6-tetra-*O*-methyl-D-glucose respectively being isolated. Until the very small particle size resin was employed no success had been achieved with this method and no other exchanger examined gave comparable results. C. V.

Dextrose and maltose contents of commercial liquid glucose. S. J. Patterson and J. L. Buchan (*Analyst*, 1960, **85**, 75—76).—The proportions of individual sugars in liquid glucose often affect the texture and storage life of confectionery and other food products, and the conventional data, viz., Baumé and "dextrose equivalent" used by manufacturers do not indicate the precise content of individual sugars. It has been shown by paper chromatography that classical methods of analysis give high results for maltose. The maltose and dextrose contents of a number of samples of liquid glucose were determined by Patterson and Savage's carbon-column method (J.S.F.A. Abstr., 1958, ii, 69) and are quoted together with the conventional data supplied by the manufacturers. A. O. Jones.

Recent developments affecting the storage of confectionery. J. G. Woodroof (*Zucker u. Süssw.*, 1960, **13**, 286—288, 290).—Nine factors are listed in approx. order of importance: humidity, temp., odour-free atm., hydrogenated fats, emulsifiers, antioxidants, humectants, zein-type coatings and mould inhibitors. The expected storage life of 18 varieties at indicated R.H. values and 0, 32, 48 and 68°F is given. The influence of thawing of confectionery after freezing is discussed and the necessity for moisture-proof packaging is stressed. C. V.

Sweetening composition. Upjohn Co. (Inventor: L. H. Macdonald) (B.P. 810,637, 4.9.57).—A sweetening composition (tablet), suitable for use in oral fluid pharmaceutical prep., comprises Cyclamate sodium and/or calcium (1), saccharin (3—10 pt., and 1 wt.-% on total composition), and tablet adjuvants. Thus, a typical cough formula of greatly reduced bitter taste contains propylene glycol (25), glycerol (40 vol.-%), phenobarbital 8, low-viscosity Na carboxymethylcellulose 10, 2-(*o*-methoxyphenyl)isopropyl(methyl)amine 4, codeine phosphate 2, Na citrate 60, saccharin sodium 40, Cyclamate sodium 5, aromatic flavour 0·2 mg. and water (to 1 c.c.). F. R. Basford.

Fermentation and Alcoholic Beverages

Analysis of spirits by vapour phase chromatography. A. Fouassin (*Rev. Ferment.*, 1959, **14**, 206—212).—The determination of alcohols, aldehydes and water in a variety of spirits using columns of Carbowax, triethanolamine and diglycerol, and hydrogen as the entraining gas is described. J. V. Russo.

Paper-chromatographic separation and determination of glycerol in wines and liqueurs. K. G. Bergner and H. Meyer (*Dtsch. Lebensmittl Rdsch.*, 1960, **56**, 49—52).—Glycerol may be determined in wine following ascending paper chromatography with water-saturated n-butanol : n-propanol (4 : 1) as mobile phase, at a temp. of 40° to 44°. Spots are located by treatment of a parallel chromatogram of wine + glycerol with ammoniacal $AgNO_3$ at 105°. For the determination, the relevant areas are cut from the paper and the glycerol is extracted with water and oxidised with periodate. The resulting formaldehyde is determined by the chromotropic acid reaction after removal of excess periodate and iodates by treatment with arsenious acid and Ag_2SO_4. Amounts of from 5 to 150 μg. of glycerol can be satisfactorily determined, in from 0·005 to 0·01 ml. of the wine. E. C. Apling.

Estimation of sulphur dioxide and sulphate ion in wines and grape juice simultaneously by iodometry and gravimetry. L. Deibner (*Rev. Ferment.*, 1959, **14**, 179—186).—The addition of SO_2 to wines and grape juice is discussed from physiological and organoleptic

viewpoints. Its chemical properties and its addition products, both inorg. and org. and precise methods for its determination are reviewed. J. V. Russo.

Determination of volatile acids in wine. H. Eschmann (*Mitt. Lebensm. Hyg., Bern*, 1959, **50**, 541—552).—The Swiss amd Austrian official steam-distillation methods are subject to considerable errors due (in the former method) to underestimation of the AcOH, and (in both) to the inclusion of lactic acid in the distillate. A shorter direct-distillation method on the lines of the Reichert–Meissl–Polenske procedure is proposed in which the above-mentioned errors are much reduced. The diluted sample (110 ml.) + NaCl (10 g.) is distilled from a 250-ml. flask fitted with an efficient globular still-head, 100 ml. of distillate being collected in 15 min. The recovery of AcOH amounts to ~96%. The lactic acid error is ~+1·5 to +4% for the range 0·3—3·6 g. per l. in the sample. The error due to distilled HCl is ~1%. Corrections for the particular apparatus in use should be determined by distillations of solutions of known content of EtOH and the acids. P. S. Arup.

Secondary products of alcoholic fermentation ; composition of fusel oils. L. Genevois and J. Baraud (*Industr. aliment. agric.*, 1959, **76**, 837—844).—Quant. relations between secondary products of alcoholic fermentation and their susceptibility to conditions in the medium are discussed. Wine, molasses and beet fusel oils were examined by vapour phase chromatography. In the first were found (besides ethanol) propanol, isopropanol (trace), butanol-2, isobutanol, methyl-3-butanol-2 (isopropyl methylcarbinol), two primary isopentanols (active and inactive amyl alcohols), and hexanol-1. In the molasses fusel, butanol-2 was absent. Beet fusels examined showed ethanol, propanol-1, isobutanol, the isopentanols, and a trace of pentanol-3. These findings are discussed, and it is claimed that the higher alcohols cannot be derived from amino-acids of the must but are the result of the activity of the yeast, representing fundamental stages in the synthesis of its own substance. (23 references.) C. L. Hinton.

Malting, brewing and allied processes, 1959. A literature survey. Editor I. A. Preece (*Inst. Brew.* 1960, 32 pp.).—The following are covered : analytical methods (88 references) ; barley and malting (214 references) ; hops and the chemistry of hops (34 references) ; brewing microbiology (83 references) ; general biochemistry (61 references) ; beer and the brewing process (14 references). E. M. J.

Improvement of brewing water with ion-exchangers. H. Lauth (*Brauwelt*, 1959, **99 B**, 1097—1102, 1158—1160).—I. The salt characteristics of brewing waters and desirable adjustments of pH through alteration of CO_3^{2-} and non-carbonate-hardness are discussed. The advantages of ion-exchange treatment for this purpose over traditional methods are indicated. By blending de-carbonated or suitably de-alkalised or de-salted water with untreated water, it is possible to approximate the composition of a desired type of brewing water such as Pilsner. Illustrations are given of changes in composition of waters of various origins effected with Lewatit CNO, Lewatit S 100 and Lewatit MIH, and of compositions attainable by blendings.

II. Mg hardness as HCO_3^- or CO_3^{2-} is best dealt with by exchange de-carbonation, as Mg unless excessive is unobjectionable. When excessive it can be removed by passing the de-carbonated water through a Ca exchanger ; by simultaneous de-carbonation and Ca exchange with Lewatit CNS ; or by complete de-alkalising followed by addition of lime or dolomite-free marble. (34 references.) C. L. Hinton.

Conversion of viscosity of worts and beers into values corresponding with a definite content of extract. P. Kolbach (*Mschr. Brauerei, wissen. Beil.*, 1960, **13**, 21—26).—Examination of Höppler's data reveals that the η of the dissolved substances (viz., the η of the wort, less that of the water) of worts from different types of malt are very similarly affected by changes in concn. Tables based on Höppler's data have accordingly been drawn up, in which values for the η of the dissolved substances and of the wort or beer are correlated with % of extract from 2 to 20. P. S. Arup.

Effect of vegetable growth substances on reproduction and metabolic rate of *Saccharomyces cerevisiae*. M. Rodríguez López (*An. Bromatología*, 1959, **11**, 461—469).—Indol-3-ylacetic acid at a concn. of 50 p.p.m. increases yeast growth by 50%. 2,4-Dichlorophenoxyacetic acid (2,4-D) reduces cell division, and at a concn. of 100 p.p.m. produces complete inhibition and lysis of the cells. 2,4-D could be used as an anti-ferment for the preservation of wine. (14 references.) E. C. Apling.

Radiation effect on permeability of yeast cells to sodium and potassium ions. K. Hsu (*U.S. at. Energy Comm.*, 1959, UCRL 9012, 102 pp.).—Movement of K^+ from the cell outwards is called diffusion ; inwards is defined as active transport. Haploid (**I**), diploid (**II**), tetraploid and hexaploid (**III**) cells of a related polyploid yeast, *Saccharomyces cerevisiae* were used. K content of cells is proportional to dry wt. while Na content is proportional to cell surface area except in the case of **I**. K leakage from the cells with distilled water is largely due to diffusion. Amount and time rate of K^+ loss is more marked in cells of larger ploidy while with higher doses the K^+ leakage is proportional to cell surface area in both control and irradiated cells. In the presence of dextrose active transport takes place, the rate of K uptake by **III** and **II** being 3 : 1, the same as the ratio of their respective vol. O_2 also initiates the active transport process but N_2 had no obvious effect on leakage or on K uptake. The order of radiosensitivity for K retentivity of cells of different ploidy does not follow the same sequence as that of lethality ; up to 107 kV did not appear to impair the active transport process or Na^+ from the cells into the distilled water. Outward movement of Na^+ could not be explained primarily as a diffusion process and it was shown that while N_2 + dextrose did not influence the Na leakage, O_2 increased it. (135 references.) C. V.

Rapid determination of the bitter substances of hops. H. J. Wellhoener and G. Irrgang (*Brauwelt*, 1959, **99, B**, 1417—1419).—The conductometric method of Hartong *et al.* (*Brauwissenschaft*, 1957, **10**, 197—198 ; *Analyt. Abstr.*, 1959, **6**, (5), No. 1928) for determination of the α-acids is described. Results obtained on 12 samples of 1958 hops show close agreement with the gravimetric method. C. L. Hinton.

Volatile constituents of beer. H. Jenard (*Brass. et Malt., Belge*, 1959, **9**, 379—381).—Contents of esters and alcohols in two types of beer before and after pasteurisation were determined by vapour phase chromatography. Distillates for analysis were dehydrated by bubbling N_2 through them and absorbing the water in a K_2CO_3 trap, the volatile compounds being condensed in a liquid air trap. J. V. Russo.

Polyphenols and the formation of cold turbidity in beer. R. Vancraenenbroeck (*Brass. et Malt., Belge*, 1959, **9**, 389—391).—The polyphenols of malt and hops were separated on ion-exchange resins and by paper chromatography and were found (especially the leuco anthocyanins) to contribute to turbidity in beer. J. V. Russo.

Nitrogenous content of barley and the cold turbidity of beer. R. Lontie (*Brass. et Malt., Belge*, 1959, **9**, 390—391).—The extraction of nitrogenous compounds from barley, the amino-acid composition of the turbidity of beer and the fractionation of the proteins and polypeptides of barley and malt are discussed. J. V. Russo.

Refractometric analyses of beer and their interpretation. W. Nebe (*Brauer u. Mälzer*, 1959, **12**, 10—14).—A revision of the Gerum–Wissner nomogram is presented, based on analyses taken from the literature of 463 beers of 3—20% original wort. The relation between alcohol, extract and original wort has been calculated strictly according to the Balling formula. C. L. Hinton.

Malting of barley. Imperial Chemical Industries Ltd., P. W. Brian and M. E. Radley (B.P. 811,374, 22.6.56).—Malting of barley is accelerated by moistening with aq. gibberellic acid prior to germination of the grain. F. R. Basford.

Worts. Brewing Patents Ltd., A. H. Cook, J. R. A. Pollock and A. D. Davis (B.P. 810,146, 5.9.56).—A continuous process for the prep. of worts comprises passing malt grist (with or without starch adjuncts) and hot water into the head of a tube or tower, keeping the contents of the latter at a temp. adequate to ensure the desired degree of enzymic conversion of the malt into sol.-extractives, then separating the residual solids from the wort at the bottom of the tube. Apparatus is figured and claimed. F. R. Basford.

[**Kaffir**] **beer.** Jabula Foods (Proprietary) Ltd. (Inventor : R. B. B. Freitag) (B.P. 811,382, 15.8.56).—A mixture of cereal medium (maize flour, mealie meal or kaffir corn meal), souring material (org. acid or acids), and diastatic malted material (kaffir corn malt flour) is cooked, then dried, to provide a product directly utilisable in the manufacture of kaffir beer. If desired, the diastatic material may be added after cooking and yeast may also be incorporated in the cooked mixture. F. R. Basford.

Fruits, Vegetables, etc.

Control of post-harvest decays of strawberries and peaches. G. R. DiMarco (*Dissert. Abstr.*, 1959, **20**, 1951).—Mycostatin (Nystatin) at 100 p.p.m. gave rather better results than Na *o*-phenylphenate at 1000 p.p.m. in decreasing decay of inoculated peaches when applied as a dip in the cooling water of a hydrocooler, and on prepackaged peaches. Na *o*-phenylphenate has been approved as a

post-harvest treatment for peaches by the Food & Drug Administration of the U.S. Treatment must be carried out within 9 hr. of harvest to be effective, or fungi penetrate the peach surface. Mycostatin was more effective than captan when applied as a dip to control post-harvest strawberry rots. Captan reduced decay, but left an objectionable residue on the fruit. Wetting strawberries is undesirable, but vac. cooling is recommended. Ripe strawberries cool faster under vac. than green ones. M. D. ANDERSON.

Utilisation of apples. V. L. S. Charley (*J. Sci. Fd Agric.*, 1960, **11,** 177—180).—Technical aspects of the utilisation of apple crops in the U.K., in Europe, U.S.A. and Canada are summarised : e.g., in England, the prep. of malic acid ; in Europe the prep. of juice and concentrate, pulps, jams, jellies, confectionery bases, etc. ; in the U.S., the prep. of sauce, slices, frozen and dried apple, juice and vinegar. In Canada three groups of products are obtained : (*a*) canned apples, apple sauce, and frozen apples which should yield to the grower their cost of production ; (*b*) products which yield half the cost of production, viz., dehydrated apples, apple juice, frozen concentrates and cider ; (*c*) such products as dried pomace and pectin. E. M. J.

Retention of *o*-phenylphenol in peel of stored oranges. W. Thode (*Dtsch. LebensmittRdsch.*, 1960, **56,** 46—49).—Results of storage tests of treated oranges covering 3, 6 and 9 weeks are reported. Fruit dipped in from 0·5% to 2·0% solutions of Na *o*-phenylphenate remained sound and unblemished. The amount of preservative penetrating into the pulp of the fruit varied from nil to 0·7 p.p.m. Storage in wrappers impregnated with *o*-phenylphenol resulted in skin damage and residues in the pulp of up to 22 p.p.m. Eleven samples of citrus products examined were free from *o*-phenylphenol and one sample of marmalade contained 0·5 p.p.m. E. C. APLING.

Ascorbic acid retention in frozen juice, segments and whole oranges. A. J. Tingleff and E. V. Miller (*Food Res.*, 1960, **25,** 145—147).—Analyses of Valencia oranges (whole peeled fruits, segments and juice) stored in freezer cups at 0 °F showed steady loss in ascorbic acid during a period of 6 months, in whatever form the material was stored. E. M. J.

Methods of analysis of soluble carbohydrates and pectic substances of citrus fruits. W. B. Sinclair and V. A. Jolliffe (*Food Res.*, 1960, **25,** 148—156).—Data show diagrammatically the scheme used to extract (80% alcohol) the alcohol-sol. fraction (**I**) of, e.g., ground citrus peel. The chief components of **I** are sol. carbohydrates and org. acids, amino-acids ; **I** also contains essential oils, pigments, flavones and biflavonoids. A diagram is given for the separation and analysis of the alcohol-insol. solids (**II**) with special reference to the fractionation of the pectic substances. **II** contain the proteins, pectic substances and other high mol. wt. compounds. E. M. J.

Recognition of potato as an adulterant in confection of sweet potato (batatada). O. A. Valenciano and M. G. Escalante (*An. Bromatología*, 1959, **11,** 471—476).—Additions of potato to sweet potato confections may be detected microscopically on the basis of differences in the starch-containing cells, and in the appearance of the gelatinised starch. The starch-containing cells of sweet potato are turgid and completely filled ; the starch is uniformly gelatinised. The cells of potato show plasmolysis—a distinct space between the cell wall and the starch cell contents, and the gelatinised starch shows radial fissures. E. C. APLING.

After-cooking darkening in potatoes. P. Muneta (*Dissert. Abstr.*, 1959, **20,** 1921—1922).—The amount of fertiliser applied to potatoes, and the addition of chelating agents with the fertiliser, did not affect the darkening of the tubers after cooking. There was no correlation of darkening with content of *o*-hydroxyphenols (determined by oxidising with polyphenol oxidase to give quinones which formed a coloured compound with aniline). Content of water-sol. Fe showed fair correlation with darkening. M. D. ANDERSON.

Determination of insecticide residues on leaves and roots of beets protected against Cleonus. F. Tafuri (*Ric. sci.*, 1960, **30,** 166—168).—Fields of beets were sprayed twice, in April and May, with aldrin, dieldrin, hexachlorocyclohexane and heptachlor, at dosages corresponding to 500, 350, 1200 and 400 g. of insecticide respectively per 10,000 sq. m. The crop was gathered in July and the residual insecticide on the combined roots and leaves determined. This was respectively 20 μg., 30 μg., 0 and 0 per kg. (10 references.) L. A. O'NEILL.

Effects of diffused light and darkness on the B-vitamin contents of germinating pulses. D. Lal Nandi (*Food Res.*, 1960, 88—96).—Thiamine, riboflavin, nicotinic acid increased in diffused light and darkness in all cases during germination, but decreased after 48-hr. germination in *Vigna catiang* and *Cicer arietinum*. Nicotinic acid contents in *Phaseolus mungo, Lens esculenta* and *V. catiang* were higher in darkness during the later period of germination. Pantothenic acid increased in diffused light and darkness in *P. mungo* and its value was higher in darkness during 96—120 hr. Folic acid decreased with germination but in *P. mungo* after decreasing, regained original value, then decreased towards the end of germination in darkness. (17 references.) E. M. J.

Determination of odour value of Mexican garlics. N. Alfonso and E. Lopez (*Z. LebensmittUntersuch.*, 1960, **111,** 410—413).—The odour of garlic is best evaluated by determination of the pyruvic acid formed when alliin, the parent substance of the odoriferous principle, allicin, is broken down by the natural enzyme of the plant. A method is described in which the pyruvic acid, after extraction from the macerated garlic, is converted into the 2,4-dinitrophenyl. hydrazone, which is then determined spectrophotometrically. Results for a number of Mexican garlics are recorded. (13 references.) C. L. HINTON.

Chao : fermented, salted and alcoholised soya cheese. C. Richard (*Industr. aliment. agric.*, 1959, **76,** 745—748).—An account is given of the nature and mode of prep. of chao, a cheese-like prep. of soya used in the Far East as a savoury adjunct to other foods. Its nutritive value is high, the average composition including : dry matter, 24% ; ash, 12% ; chlorides (as NaCl), 11% ; protein, 7% ; fatty matter, 3·5%. (18 references.) C. L. HINTON.

Non-alcoholic beverages

Production of mahewu. F. Schweigart, W. E. L. van Bergen, S. G. Wiechers and J. P. de Wit (*Counc. sci. industr. Res. Pretoria, S. Africa*, 1960, Rep. No. 167, N.N.R.I. Bull. No. 3, 29 pp.).—The prep. of this non-alcoholic beverage of the Bantu is described in detail especially with regard to fermentation processes, during which mainly lactic acid is formed. Drying, prep. of paste, nutritional value and enrichment are discussed. E. M. J.

[A] **Utilisation of citrus fruit.** E. H. G. Smith and D. E. Kay. [B] **Addendum.** J. W. Seymour (*J. Sci. Fd Agric.*, 1960, **11,** 181—188, 189—191).—[A] The production of major and some minor citrus products are briefly reviewed in generalised terms, covering : juice products, canned segments, marmalade, fermented products, citric acid production, by-products, e.g., essential oils, dried citrus pulp and molasses, pectin, candied peels, citrus seed oil and canning syrup. (18 references.)

[B] A survey of pre-war and present-day production, popularity of various products and economic aspects is presented. E.M.J.

Citrus fruit concentrate. Union Carbide Corp. (B.P. 811,798, 25.5.56. U.S., 26.1.56).—A citrus fruit juice concentrate of improved stability is obtained by removing the flavedo layer from citrus fruit, submitting the fruit (containing the albedo layer and tissue enclosing the juice substantially intact) to enough pressure to rupture it and to remove the juice without crushing the seed ; separately compressing the albedo and juice cell tissue to remove the albedo juice ; adding the latter (1—5%) to the fruit juice ; and (before or after said addition) removing water from the fruit juice in successive freeze dehydration stages (at 5°F). F. R. BASFORD.

Tea, coffee, cocoa

Phenolic substances of manufactured tea. VII. Preparation of individual flavanols. VIII. Enzymic oxidations of polyphenolic mixtures. E. A. H. Roberts and M. Myers (*J. Sci. Fd Agric.*, 1960, **11,** 153—157, 158—163 ; cf. J.S.F.A. Abstr., 1959, i, 334).—VII. Methods which employ Craig counter-current distributions and column chromatography on Magnesol–Celite and silica gel are described for the prep. from dried green tea-leaf of (−)-epigallo-catechin, (+) gallocatechin, (−)-epigallocatechin gallate and (−)-epicatechin gallate. The isolation of (+)-gallocatechin confirms earlier predictions that this isomer, and not the racemate, would be found in unprocessed tea-leaf. (25 references.)

VIII. When a mixture of substrates is acted on by tea oxidase, the substrate of lower rH is oxidised preferentially. Oxidations in tea fermentations are mainly concerned with those of the gallocatechins. Mixed dimers are produced only when two substrates have approx. the same rH values, as with a mixture of (−)-epigallocatechin and its gallate. Gallic acid and theogallin may undergo coupled oxidations. The substance Q is considered to be a mixture of small amounts of three substances all originating from gallic acid by coupled oxidation. The oxidation products of theogallin may be present as minor constituents of the thearubigin complex. Theaflavins, bisflavanols and thearubigins undergo coupled oxidations with the catechins as carriers. The thearubigin fraction undergoes continual change during fermentation. (12 references.) E. M. J.

Milk, Dairy Products, Eggs

Effect of season and stage of lactation on yield and composition of milk in two herds of University College of Wales, Aberystwyth. M. S. Bayoumi (*Indian J. Dairy Sci.*, 1959, **12**, 87—99).—Data on the milk yield and its butter fat, solids-not-fat and total solids contents in a Dairy Shorthorn herd and an Ayrshire herd were collected over 2 years. The correlations between the milk yield and its constituents are calculated and discussed. The season of the year and the stage of lactation have considerable effect on the yields and correlations. (16 references.) S. G. AYERST.

Vitamin A in milk. X. Factors influencing vitamin A and carotene contents of cow colostrum. K. M. Narayanan and C. P. Anantakrishnan (*Indian J. Dairy Sci.*, 1959, **12**, 106—116).—Colostrum taken from cross-bred, Gir and Red Sindhi cows was analysed and the average yields, fat %, carotene (**I**) and vitamin A (**II**) contents were determined. Marked individual variations were found in the first post-partum secretion. The difference in total output and in concn. of **I** and **II** in the different breeds was statistically significant. Lactation no. and length of non-lactating period had no effect on the contents of **I** and **II**. The first colostrum from animals calving in the monsoon season was richer in **I** and **II** than that from animals calving in the hot or cold seasons. (28 references.) S. G. AYERST.

Effect of temperature on creaming of buffalo milk. I. S. El-Hagarawy and S. E. S. E. Rakshy (*Indian J. Dairy Sci.*, 1959, **12**, 117—120).—The creaming behaviour of both cow and buffalo milk at different temp. was investigated. Creaming in cow milk reached its max. at 40°F and its lowest at 100°F. There was no big difference in the creaming tendency at 77°F and at 100°F. On the contrary, creaming in buffalo milk reached its max. at 100°F and was lowest at 40°F. S. G. AYERST.

Tocopherol content of Indian butter and its use in detecting adulteration of butter-fat. D. J. Nazir and N. G. Magar (*Indian J. Dairy Sci.*, 1959, **12**, 125—132).—Tocopherol contents of a few samples of Indian butter and butter-fat were estimated by the $FeCl_3$–α, α′-dipyridyl and PMA methods, after the removal of interfering substances by Dunford's method. Corrections were applied for carotenoids. Adulteration of butter by oils rich in tocopherol could be detected at a level of 10% and in some cases at a level of 5%. (22 references.) S. G. AYERST.

Co-vitamin studies. IV. Stability of tocopherol, carotene and vitamin A in ghee during storage. K. M. Narayanan and C. P. Anantakrishnan (*Indian J. Dairy Sci.*, 1959, **12**, 133—138).—Samples of ghee obtained by the cream, creamery and *desi* methods, from both cow and buffalo milk, were stored at 37° for 6 months and analysed at intervals to find the peroxide value (**I**), carotene (**II**), vitamin A (**III**) and tocopherol (**IV**) contents. The method of prep. had no effect on the initial value of **II**, **III** and **IV**, but during storage the vitamins were found to be less stable and the values of **I** were higher in *desi* ghee than in cream or creamery ghee. (14 references.) S. G. AYERST.

Application of paper chromatography to differentiate ghee from other fats. I. Behaviour of unsaponifiable matter on chromatograms. B. V. Ramachandra and N. N. Dastur (*Indian J. Dairy Sci.*, 1959, **12**, 139—148).—Samples of unsaponifiable matter from ghee and other fats were spotted on to Whatman no. 1 filter paper and irrigated. 28 solvents were tested and finally methyl alcohol–light petroleum–water, 80 : 10 : 10, was adopted. Under u.v. light adulteration of ghee by more than 5% vanaspati could be detected by intense fluorescence. 5% of animal body fats made the ghee spot immobile and brightly fluorescent, whereas pure ghee spots moved some distance. Edible vegetable oils and cow, buffalo, goat and sheep ghee behaved similarly and could not be differentiated. S. G. AYERST.

Turbidimetric micro-determination of magnesium in milk. J. R. Marier and M. Boulet (*J. Dairy Sci.*, 1959, **42**, 981—988).—A simple and convenient direct method is described for the determination of Mg in milk. After removal of Ca as oxalate, 5—40 μg. of Mg can be determined by measuring the turbidity produced by the K salt of erucic acid in the presence of K oxalate. Results obtained agree within ±3·3% with those obtained on ashed samples. S. C. JOLLY.

Composition of ultrafiltrates from milk heated at 80° to 230°F in relation to heat stability. D. Rose and H. Tessier (*J. Dairy Sci.*, 1959, **42**, 969—980).—In milk at 230°F, the amount of Ca and phosphate passing into the ultrafiltrate was ~50 and 82% respectively of that passing at 80°F; the concn. of H^+ in the hotter filtrate was more than twice that in the cooler. These changes in the inorg composition of milk can be explained entirely on the basis of changing solubility and composition of the insol. Ca phosphates. The dissociation of Ca citrate was unaffected by temp. The changes induced by heat were not correlated with the heat stability of the milk. S. C. JOLLY.

Detection of intermediate compounds in the early stages of browning reaction in milk products. M. Keeney and R. Bassette (*J. Dairy Sci.*, 1959, **42**, 945—960).—A spectrophotometric method, based on reaction with 2-thiobarbituric acid, is described for the determination of 5-hydroxymethylfurfural (**I**) and other furans. For converting early intermediates of the browning reaction to **I**, and so providing a sensitive method of detecting early symptoms of the reaction, a selective digestion with oxalic acid is used. Addition of **I** to condensed milk before spray-drying caused off-flavours in the product. The development of "cereal-stale" type of flavour in skim-milk powder is apparently associated with browning. Some commercial "instantising" processes promote the browning reaction. S. C. JOLLY.

Influence of added 1-monoglycerides on the surface tension of milk. A. H. Duthie and R. G. Jensen (*J. Dairy Sci.*, 1959, **42**, 863).—The addition of two drops of 1-mono-olein (92), 1-monolaurin (84), 1-monopalmitin (94) and 1-monostearin (70% purity) to 100 ml. of composite Holstein milk reduced the surface tension on average by 6·3, 5·2, 0·9 and 0·4 dynes per cm. respectively. Even very small amounts (0·2 mM) of monolaurin and mono-olein, added in ethanolic solution, noticeably reduced surface tension. Since 2·0 mM of monoglyceride per 100 g. of fat has been detected in very rancid milk, sufficient surface-active monoglycerides may be present in milk in various stages of lipolysis to affect foam and whipping, the fat-globule surface, and other surface and interfacial phenomena. S. C. JOLLY.

Influence of bacterial interaction on resazurin reduction times. V. W. Greene and R. M. Jamison (*J. Dairy Sci.*, 1959, **42**, 1099—1100).—Typical bacterial species such as *Streptococcus lactis*, *Pseudomonas fluorescens* and *Bacillus subtilis*, isolated from raw milk, affected each other's ability to reduce resazurin when grown in reconstituted skim milk powder. Reduction was accelerated, sometimes significantly, when combinations of cultures were used. The strong reducing action of *Str. lactis* overshadowed any interactions between this culture and the others; the most marked interaction was between *B. subtilis* and *Ps. fluorescens* in milk of good quality. S. C. JOLLY.

Staphylococci in milk. G. Obiger (*Milchwissenschaft*, 1960, **15**, 107—113).—A review. (30 references.) C. V.

Growth of staphylococci in condensed skim milk. E. George, jun., J. C. Olson, jun., J. J. Jezeski and S. T. Coulter (*J. Dairy Sci.*, 1959, **42**, 816—823).—The growth of two staphylococcal cultures was unaffected by preheating skim milk at 165° or 185°F for 30 min. before condensing; the growth of one was retarded by preheating at 150°F. Optimum growth, or nearly so, occurred in condensed milk at 90° to 113°F with solids concn. of ~30 to 50%; three of six cultures grew, although slowly, in 40% milk solids at 116°F, and one grew at 118°F. Under conditions simulating vacuum distillation, growth was less than under normal atm. pressure, but sufficiently rapid to preclude the practical use of reduced pressure to control growth. S. C. JOLLY.

Detection of *Staphylococcus aureus* in non-fat dry milk by fluorescent-antibody technique. P. B. Smith (*Dissert. Abstr.*, 1959, **20**, 1969).—Use of the indirect fluorescent-antibody technique on ~75 strains of *Staphylococcus aureus* enabled 83% of the strains to be specifically identified in presence of morphologically similar and dissimilar contaminants. Interference was only caused by cells of *Bacillus cereus*. The method was also used for the quant. detection of *S. aureus* at a concn. of $3{\cdot}6 \times 10^6$ cells per ml. and at greater dilutions. The prep. of broad-spectrum antisera and the testing technique are described. A. M. SPRATT.

Sterilisation of milk by ionising radiations. M. Worseck (*Milchwissenschaft*, 1960, **15**, 114—119).—A review with special reference to the flavour that may result. (50 references.) C. V.

Survey of radioactivity of milk products. I. A. Miserez (*Mitt. Lebensm. Hyg., Bern*, 1959, **50**, 508—522).—Records for 1958—59 of the total β-radioactivity of milk (determined on the ash) show, during pasture-feeding, considerable variations which are directly correlated with the amount and radioactivity of the rainfall. The average activity of the insol. oxalates derived from the milk constitutes ~80% of the activity due to nuclear outfall; the difference between the total and insol. oxalate activity is represented by that naturally due to ^{40}K. The ^{40}K activity remains fairly constant throughout the season for the mixed milk of large herds, but shows considerable variations (sometimes simulating contamination) for that of very small herds. No unexpected differences have been observed between contamination in mountainous and lowland districts of Switzerland. P. S. ARUP.

Examination of casein preparations by paper electrophoresis. M. T. Sode-Mogensen and E. Lahav (*Lab. Practice*, 1960, **9**, 21—28, 34).—Good separations of α-, β- and γ-casein are obtained by constant-voltage electrophoresis (4·7 V/cm.) on Whatman 3 MM paper, using a veronal buffer as electrolyte, in 16 hr. at 4°. The separated components are located with Naphthalene black 12 B 200. J.A.C. ABSTR.

Size memory of casein colloid particles. W. L. Choate, F. A. Heckman and T. F. Ford (*J. Dairy Sci.*, 1959, **42**, 761—766).—When dialysed against Na barbiturate buffer, washed Ca caseinate fractions of varying size were converted to identical solutions of Na caseinate, as indicated by ultracentrifugal pattern. When redialysed against skim milk, these Na caseinates aggregated to a size range dependent on that of the original micelles from which the Na caseinates were made. Reaggregation of casein is dependent on factors other than Ca^{2+} concn. alone. S. C. JOLLY.

[A] **Fat content of milk.** F. Custot. [B] J. Pien (*Ann. Falsif., Paris*, 1959, **52**, 460—463, 464—469).—[A] The latest French legal requirements for the fat content of "standard" milk (26 g./l.) are discussed and criticised and compared with requirements in other countries.

[B] Economic problems posed by the lowering of the fat content requirements for "standard" milk, particularly during a period of excessive drought, are discussed. J. V. RUSSO.

Industrial refrigeration and alterations in milk and dairy products J. Moreno-Calvo (*Industr. aliment. agric.*, 1959, **76**, 721—727, 875—888).—The physico-chemical structure, biochemical qualities and bacterial contamination of milk, butter, cream, margarine, cheese and ice cream are reviewed, and a wide survey of the literature is presented on the changes undergone in refrigeration, freezing and thawing of both raw and pasteurised products. The results of unpublished work by the author on chemical, physico-chemical and organoleptic changes occurring in the storage at temp. from −19 to −5° of fresh milk quick-frozen at −37° are briefly described. (386 references.) C. L. HINTON.

One-dip nuclear staining procedure for direct microscopical examination of milk, milk products and other foods. C. W. Anderson (*J. Milk Tech.*, 1960, **23**, No. 2, 37—39).—The advantages of a $CHCl_3$–MeOH–Azure A defatting–fixing solution are compared with the use of polychrome methylene blue. Staining of milk solids and bacteria is carried out in 1 min. but will be longer with dried egg (10% solution) or diluted frozen egg. Longer staining even up to 24 hr. is neither more, or less, effective. (10 references.) C. V.

Double and triple tube heat exchangers. Anon. (*Milk Dealer*, 1959—60, **48**, No. 9, 43—44, 61).—A brief review of the effectiveness of this tube-within-a-tube type of heat exchanger with special reference to space saving. C. V.

Shelf-life of creamed cottage cheese; extension with sorbic acid. J. Geninder (*Milk Dealer*, 1958—59, **48**, No. 4, 44—45, 133—134, 140).—Addition of sorbic acid, Na- or K-sorbate (0·05—0·1%) extended the life of non-acid-washed creamed cottage cheese by 30 days; yeast and mould growth were inhibited. It is stressed that off-flavour and slime development are apparent before visual growth or gas development are noted. In acid-washed cheese (pH 4·0—4·8), the life was extended to 52 days. C. V.

Fermented milk products. K. Mishima (B.P. 809,598, 6.6.55).—A yoghurt-like product is obtained by fermenting the milk by inoculation with a first quantity of yoghurt bacteria and with a saccharomyces yeast (*S. fragilis, S. ellipsoideus*, etc.) effective to produce alcohol and aroma. Mucilage, e.g., methylcellulose or agar agar, and a natural fruit product are added and the pH of the mixture is adjusted to 5—5·5. A quantity of unfermented milk is then added, the mixture is pasteurised by heat, then cooled, and a lactic acid fermentation is effected by inoculation with a further quantity of yoghurt bacteria, e.g., *Lactobacillus bulgaricum, Bact. acidophilum*, etc. J. M. JACOBS.

Edible Oils and Fats

Differentiation of refined from non-refined cold-pressed olive oils. E. Lauber (*Mitt. Lebensm. Hyg., Bern*, 1959, **50**, 553—566).—An apparatus specially designed for determining the sp. electrical resistance of olive oil is described. Samples showing sp. resistances of <1000 megohms, acid value of >1 (range 1—6), and a well-defined extinction peak at 670 mμ can very probably be classed as cold-pressed. Results obtained for 34 samples are discussed in relation to their commercial description, taste and appearance. P. S. ARUP.

Lipase and lipoxidase in olives. C. Cantarelli (*Olii min.*, 1960, **37**, 2—7).—The occurrence of lipase in the endosperm, mesocarp, vegetation water and oil of olives has been examined by observing the lipolytic activity of various extracts for tributyrin and other substrates. The prep. from the endosperm and mesocarp showed similar activity for tributyrin, which was at a max. at pH 6·5. The former was comparatively more active for methyl oleate and the latter for methyl linoleate. Considerable activity was shown by the vegetation water but not by the oil. Lipoxidase activity of similar extracts was examined by a method involving the oxidation of linoleate and the bleaching of β-carotene by the product (cut-plate method). Lipoxidase appeared to be present exclusively in the endosperm. The mesocarp, in contrast, contained compounds, e.g., polyphenols, which inhibited the action of lipoxidase. The oxidation of olive oil or fatty acid esters when kept over the vegetation water was reduced as compared with pure H_2O. (31 references.) L. A. O'NEILL.

Packaging, transportation and storage of edible vegetable oils. M. Prasad and P. B. Mathur (*J. sci. industr. Res.*, 1959, **18A**, 571—575).—The storage lives of eight edible vegetable oils in colourless and deep green bottles were determined at normal temp. (71—93°F) and in the range (138—142°F) likely to be encountered during transport in the tropics. The oils were examined at intervals for acidity, peroxide value, and I val. A positive correlation was found between red/yellow pigment ratios and storage lives; a high ratio offset the effects of degree of saturation and colour of the container. Deterioration is much more rapid in the higher-temp. range, and may not be controlled by adjusting the pigment ratio. Refrigerated transport is recommended for the tropics. (10 references.) A. M. SPRATT.

Chromatography of the sterols, and its application for detection of animal and vegetable fat mixtures. J. W. C. Peereboom and J. B. Roos (*Fette Seif. Anstrichm.*, 1960, **62**, 91—100).—The fat mixture is saponified with 40% ethanolic KOH. and the sterols are precipitated by the addition of 1% digitonin solution. The free sterols are then separated by ascending chromatography on paper impregnated with paraffin, with 84% acetic acid as the mobile phase. The sterols are thus separated from the phytosterols and are located by spraying the paper with phosphomolybdic acid. Small quantities of vegetable fat in animal fat or vice versa can be determined by this technique. G. R. WHALLEY.

Bleachability of Nigerian palm oil. G. R. Ames, W. D. Raymond and J. B. Ward (*J. Sci. Fd Agric.*, 1960, **11**, 194—202).—Good bleachability of palm oil from Nigerian palm fruit may be obtained if sufficient care is taken in processing. Native-produced oil is difficult to bleach owing to oxidation of the oil by lipoxidases, occurring when bruised unsterilised fruit is left for various periods before extraction of the oil. Other factors are: atm. oxidation catalysed by Fe may cause a further deterioration in the bleachability of the oil during processing; the mixing of oils having good and bad bleachability; enzymic oxidation resulting in formation of yellow pigments less readily absorbed on Fuller's earth, and less readily degraded by heat than the original carotenoids. An oil (e.g., Nigerian) having a high carotene content is more liable to deteriorate than an oil (e.g., Malayan) with a low carotene content. E. M. J.

Permeability of fat products to moisture. W. Landmann, N. V. Lovegren and R. O. Feuge (*J. Amer. Oil Chem. Soc.*, 1960, **37**, 1—4).—Permeability constants (P) at 80 and 37·4°F of enrobing and other confectionery fats have been determined on films from 1—3 mm. using the cup method. With cocoa butter, permeability was greater with moderate increases in film thickness and was considerably affected by the polymorphic state of the fat. Thus, values of P for quickly chilled and tempered films were 310×10^{-12} and 33×10^{-12} respectively under similar test conditions. The addition of 40% liquid cottonseed oil to the hydrogenated oil increased P approx. 300-fold. High R.H. had little effect on cocoa butter or hydrogenated cottonseed oil, but a much greater permeability occurred with chocolate liquor and sweet milk enrobing chocolate. P. M. KINGSTON.

Hydrogenation of fatty oils with palladium catalyst. III. Hydrogenation of fatty oils for shortening stock. M. Zajcew (*J. Amer. Oil Chem. Soc.*, 1960, **37**, 11—14).—Hydrogenations of soya-bean oil, cottonseed oil and a 7 : 3 mixture of the two have been made, varying pressure, temp., agitation, concn. and feedstock I no. and using Pd/C of different activity. Selectivity and control of *trans* isomer content appeared good and, under the right conditions (generally mild), several shortening stocks were obtained having good plastic properties. (30 references.) P. M. KINGSTON.

Isomerisation of fats during hydrogenation: metabolism of component fatty acids. R. R. Allen and P. V. Johnston (*J. Amer. Oil*

Chem. Soc., 1960, **37**, 16—18).—The extent of positional and geometrical isomerisation which occurs during catalytic hydrogenation of edible oils and fats is discussed, together with the metabolic fate of the isomers. (20 references.) P. M. KINGSTON.

Calculation of distribution of saturated and unsaturated acyl groups in fats from pancreatic lipase hydrolysis data. R. J. Vander Wal (*J. Amer. Oil Chem. Soc.*, 1960, **37**, 18—20).—A method is proposed for calculating proportions of glyceride types and isomers in fats where C_{16-18} chains predominate. Examples are given for kokum and cocoa butters, groundnut and soya-bean oils, and pig, beef and rat fats. P. M. KINGSTON.

Distillation method for determination of malonaldehyde in rancid foods. B. G. Tarladgis, B. M. Watts, M. T. Younathan and L. Dugan, jun. (*J. Amer. Oil Chem. Soc.*, 1960, **37**, 44—48).—A simplified Sidwell distillation technique is described for meat and compared with Turner's extraction method. A 10% aq. meat slurry (adjusted to pH 1·5 with HCl) is boiled in a Kjeldahl flask until half the water has distilled. Aliquots treated with 0·02M-2-thiobarbituric acid (TBA) in 90% glacial acetic acid are then compared spectrophotometrically with standard curves using a 0·001M aq. solution of 1,1,3,3-tetraethoxypropane (TEP). The effect of varying pH, heating time and recovered distillate was examined in relation to the TEP standard. Less fat oxidation occurs during the test itself than with other techniques. High correlation of TBA no. with rancid odour is also established although malonaldehyde only plays a small part in the basic odour. (29 references.) P. M. KINGSTON.

Antioxidant activities of the tocopherols. II. Influences of substrate, temperature and level of oxidation. C. H. Lea (*J. Sci. Fd Agric.*, 1960, **11**, 212—218; cf. J.S.F.A. Abstr., 1960, i, 102).—Activities were compared at 60° and at 37° in distilled methyl esters of cottonseed, linseed and cod-liver oil fatty acids containing added oxidised ester as "starter." In the linoleate (cottonseed) system the γ- and δ-compounds were the most and the α-, ζ- and ϵ-compounds the least effective in extending the induction period. In the polyunsaturated (**I**) (linseed, cod-liver oil) systems γ-tocopherol was good, α-tocopherol was at the top of the activity sequence and δ at the bottom. In **I** substrates antioxidant activity increased with increasing nuclear methylation especially at the lower temp. and the lower peroxide level. E. M. J.

Mycological synthesis of fat from whey. II. Comparison of shaken and stationary cultures using selected moulds. P. Wix and M. Woodbine (*J. appl. Bact.*, 1959, **22**, 175—183).—*Aspergillus ustus* (**I**), *Penicillium oxalicum* (**II**), *P. frequentans* (**III**) and *P. notatum* (**IV**) were studied. **I** and **II** showed highest lactose utilisation and felt wt. in shaken cultures; **III** and **IV** gave slightly superior findings in stationary ones. Addition of N to cultures of **I** led to increased fat production but **III** did not respond to this. With NH_4NO_3 **I** utilised 96% of lactose forming ~17 g. mycelial felt per l. of whey. The felt contained 13% of protein and 28% of fat. (21 references.) C. V.

Examination of a "synthetic" butter fat. E. Hanssen, W. Sturm and H.-J. von Drachenfels (*Z. LebensmittUntersuch.*, 1960, **111**, 381—393).—A so-called synthetic butter-fat, Bolzella fat, said to be made by transformation of beef tallow, was compared with genuine butter-fat and with beef tallow. The Bolzella fat simulated butter-fat closely in most chemical and some physical characteristics, but showed a deficiency in phosphatides and excess of lauric acid, and gave negative vitamin A reaction in the unsaponifiable matter. In luminescence and crystallographic formation it differed markedly from butter-fat. Organoleptically, both as fat and after incorporation of water, it was unsatisfactory as an alternative to butter, although bakery products prepared with it had a weak butter flavour. Its alleged origin from beef tallow could neither be confirmed nor refuted with certainty. Deteriorated cold-store butter is suggested as a possible source material. (69 references.) C. L. HINTON.

Carotene, vitamin A and vitamin E content of yellow beef fat. A. Mirna (*Z. LebensmittUntersuch.*, 1960, **111**, 393—402).—Chromatographic methods used in determining the carotene (α-, β- and γ-), vitamin-A and vitamin-E contents of 24 beef fats (suet and brisket fats) are described. Carotene contents varied over wide limits (from 24 to 690 μg./100 g. in quite fresh fats), the β-form making the major contribution, averaging 65% of the total. Contents of vitamin A and vitamin E were relatively small, averaging 1·7 i.u./g. and 1·3 mg./100 g. respectively. No marked differences were found between suet and brisket fat. The yellow coloration of beef fat consists largely of biologically valuable substances, principally derived from the feed. (18 references.) C. L. HINTON.

β-Monoester content of commercial monoglycerides after prolonged storage. L. Hartman (*J. Sci. Fd Agric.*, 1960, **11**, 191—194).—The existence of β-monoglycerides in commercial prep. stored 1·5—5 years was re-examined; "total" and β-monoglyceride were determined by isomerisation with perchloric acid; 5—9% of the total was β-monoglyceride. These findings were in agreement with those of Brokaw *et al.* (5—8%) but differed from those of other workers who considered that β-monoester present in freshly prepared products disappeared on storage. E. M. J.

Liquid shortening. T. Hedley & Co. Ltd. (B.P. 810,277, 26.10.56. U.S., 28.10.55).—An opaque to translucent liquid shortening consists essentially of a liquid glyceride vehicle and, in suspension therein, 2—10% of finely divided solid material including 1—2·5% by wt. of the shortening of a solid "high ratio" emulsifier, the balance of the solid material consisting of saturated glycerides at least 80% of which are in the form of β-phase crystals (cf. following abstract), the shortening having an increase in solids content of $>20\%$ of the solids content at 100°F when the shortening is cooled from 100°F to 60°F, the shortening being pourable at a temp. as low as 60°F. E. ENOS JONES.

Stable pourable oleaginous suspensions. T. Hedley & Co. Ltd. (B.P. 810,278, 26.10.56. U.S., 28.10.55).—A pourable oleaginous suspension of finely divided, normally solid, glycerides in a normally liquid glyceride vehicle, the solid glycerides being capable of existing in a stable β-crystalline phase and comprising 1—20% by wt. of the suspension, is made by forming a mixture of the solid glycerides and liquid glyceride vehicle at a temp. sufficiently high to melt all the solid glycerides, cooling the mixture to a temp. below the α melting temp. of the solid glycerides in >3 min., and tempering the mixture to convert non-β-crystals to a β-phase to such an extent that at least 60% of the solid glycerides are in a β-phase. E. ENOS JONES.

Simultaneous production of refined shark liver oil and vitamin A concentrates with recovery of other therapeutically active by-products. S. Mahdihassan, M. Moinuddin, S. Maqsood Ali and S. Abdul Haq (B.P. 810,643, 16.5.57).—A process for the prep. of vitamin A concentrate and refined shark liver oil containing vitamin A from crude shark liver oil, comprises allowing the crude oil to separate into solid and liquid phases, refining the oil phase to obtain refined oil containing vitamin A, saponifying the solid (stearin) phase, solvent extracting the unsaponified matter, evaporating off of the solvent and separating cholesterol present in the extract to yield vitamin A concentrate. E. ENOS JONES.

Fatty materials. Unilever Ltd. (Inventor: C. B. Cox) (B.P. 810,006, 6.4.56).—Solid discrete particles of a fatty material are made by forcing the material in liquid form through an orifice under conditions of substantially streamline flow to form a liquid jet, subjecting the jet to vibrations of a sonic frequency such as to cause the jet to separate into discrete liquid particles, allowing the particles to travel for a distance at least that required for the particles to solidify and collecting the particles. The frequency is between $u/6d$ and $u/\pi d$ where u is the velocity of the stream in mm. per sec., and d the diameter of the orifice in mm. E. ENOS JONES.

Margarine. R. Knollenberg, C.-G. Hahn and K. von Holdt (Schröder & Co.) (B.P. 810,275, 13.6.57).—An improved process for the manufacture of margarine in which the resting stage is omitted, comprises causing a margarine emulsion to travel with vigorous stirring through a tube-like cooler containing a rotatable shaft provided with scraper blades, the rate of flow of the emulsion being 1200—1600 (instead of the usual 1800) litres per hr. per sq. m. of internal cooling surface. F. R. BASFORD.

Meat and Poultry

Quantitative determination of the tranquilliser Disquel in animal tissues. R. Adjarian (*Food Res.*, **25**, 113—119).—The coloured reaction of Disquel deriv. (cf. phenothiazine) with H_2SO_4 was used as a base of the colorimetric method. The findings on rate of absorption from the site of injection in rabbits and the necessity of a new route of administration are discussed. The possibility of high concn. of Disquel at the site of injection (intramuscular), e.g., as in beef cattle for slaughter, would mean the loss of large pieces of meat, not suitable for human consumption. (14 references.) E. M. J.

Factors affecting the water retention of beef. II. Variation in pH determinants among eight muscles. C. E. Swift, M. D. Berman and C. Lockett (*Food Technol.*, 1960, **14**, 74—79; cf. J.S.F.A. Abstr., 1960, i, 94).—A comparative study was made of the glycogen and protein content, pH and buffering capacity of eight different

muscles from each of seven bovine animals. Glycogen and buffering capacity varied considerably, variation following patterns shown to be characteristic of the series of muscles with regard to pH, water retention and other properties. The content of residual glycogen was largest in muscles attaining the lowest ultimate pH values. Buffering capacity tended to be largest in muscles in which low pH is a characteristic. An explanation is offered, viz., that ultimate pH varied with the amount of glycolysis occurring prior to disappearance of adenosinetriphosphate and that the amount of glycolysis occurring during this interval was affected by variations in rates. (16 references.) E. M. J.

Behaviour of meat pigments in solutions. M. Glidden, M. Mangel, K. Singleton and M. Stone (*Food Res.*, 1960, **25,** 127—138).—The behaviour of meat pigment extracts after addition of H_2S gas, aq. H_2S, Na hydrosulphite, cysteine, glutathione and 1-amino-2-naphthol-4-sulphonic acid, respectively, was studied. The S-containing pigment described by Keilin may be present in mixed pigment solutions. This pigment seems to persist under conditions which bring about conversion of pigments into metmyoglobin, but appears to be completely converted into cyanmetmyoglobin. E. M. J.

Effect of addition of antioxidants to frozen ground beef. H. M. Caldwell, M. A. Glidden, G. G. Kelley and M. Mangel (*Food Res.*, 1960, **25,** 139—144).—Ascorbic acid (**I**) used alone or in combination with each of the following: nordihydroguaiaretic acid (**II**), ethylenediaminetetra-acetic acid, Na citrate or dihydroxyquercitin maintained a good colour in thawed samples during frozen storage for 6 weeks. Antioxidants other than **I** failed to protect the colour of the meat; **II** contributed an undesirable flavour. (11 references.) E. M. J.

Action of the "nitrite-pH-redox" conjunction on the colour and the preservation of charcuterie products. J. Maillet and M. Henry (*Industr. aliment. agric.*, 1959, **76,** 709—713).—It is shown that the employment of a mixture of NaCl, KNO_3 and sucrose in charcuterie products and brines, customary in France, by reduction of nitrate to nitrite and oxidation of sucrose to lactic acid, leads to a pH (5·4 to 6) at which the NO_2^- is in a condition of mild instability favourable to its bacteriostatic action, as well as to other desirable effects, including its reducing action on myoglobin pigment, an antioxidative protection of the fat, and a denaturing effect on muscle protein. (14 references.) C. L. HINTON.

Consumer quality of selected muscles of raw and cooked pork. O. M. Batcher and E. H. Dawson (*Food Technol.*, 1960, **14,** 69—73).—The degree of marbling in raw *longissimus dorsi* muscle between the 10th and 11th rib shows promise as an indicator of juiciness and tenderness of the cooked meat. Greater variations in fat content, colour and tenderness were noted among muscles from some carcasses than from others. (14 references.) E. M. J.

Supplementation of chick diets with vitamin E to improve meat quality. B. Laksesvela (*J. Sci. Fd Agric.*, 1960, **11,** 128—133).—Chicks were fed diets (i) containing 6 or 15% of prepared herring meal containing highly unsaturated fat (normally considered undesirable for production of good flavour in the meat), (ii) commercial herring meal or (iii) all vegetable. Addition of 36·7 mg. of *d*-α-tocopheryl acetate per kg. of diet improved the palatability of the chicken meat in general, significantly in the case of diets (i) and in (ii) with 15% of herring meal. Administration of a large dose of vitamin E during the first week of life enhanced growth rate in early stages, and, given in the first week or last five days, enhanced the organoleptic effect. (12 references.) E. M. J.

Biochemistry of chicken muscle as related to rigor mortis and tenderisation. D. de Fremery and M. F. Pool (*Food Res.*, 1960, **25,** 73—87).—In chickens 10 to 16 weeks old muscle at room temp. passes into rigor mortis 2—4½ hr. post mortem; ultimate pH is 5·8—5·9 and initial concn. of adenosinetriphosphate (ATP) is 4·8 mg./g. of muscle. Every treatment (e.g., excising the muscles pre-rigor, mechanical beating, post-mortem environmental temp., freezing and thawing of fresh muscle, exhaustive electrical stimulation, electron irradiation and lethal injections of Na monobromoacetate) that resulted in more rapid loss of ATP, more rapid drop of pH and loss of glycogen induced increased muscle toughness. Relative toughness of cooked muscle increases with increasing rate of onset of rigor mortis. (30 references.) E. M. J.

Fish

Determination of chlortetracycline in tissues. I. Improved pad-plate method. II. Improved cylinder-plate method. T. Tomiyama, A. Tsuda and Y. Yone (*Food Res.*, 1960, **25,** 97—106, 106—112).—I. The method was improved with respect to its sensitivity, accuracy and recovery of chlortetracycline from tissue. The routine assay method was adapted by adjusting the pH of the medium to 5·6, using *Bacillus cereus* No. 213 instead of *B. cereus* No. 5, holding the pad plates in the refrigerator for 1 hr., using a mixture of K citrate (pH 5·6) and acetone as extractant (recovery is not affected by kind, freshness and chlortetracycline content of, e.g., fish flesh examined), etc. (24 references.)

II. A mixture of acetone and citrate buffer was used for the extraction of chlortetracycline from samples and for diluting the standard chlortetracycline solution. Recovery was 95% in several kinds of fish flesh and a sensitivity of 0·002 μg./ml. E. M. J.

Spices, Flavours, etc.

Characterisation of vanilla and other plant extracts by paper chromatography. H. P. Burchfield and E. A. Prill (*Contr. Boyce Thompson Inst.*, 1959, **20,** 251—263).—A method based on chromatographing on Whatman No. 1 with a solvent containing KOH, NaBr and ethanol, with a solvent containing isopropanol, aq. NH_3, NaBr and water, and a solvent containing acetic acid, HCl and water is described. When viewed under u.v. light, fluorescent patterns were seen that were characteristic of the plant extracts, and the intensity of fluorescence was proportional to concn. E. G. BRICKELL.

Qualitative micro-investigations of carbonyl compounds by the radial chromatography of their hydrazones. L. Peyron (*Fette Seif. Anstrichm.*, 1960, **62,** 114—115).—Plant material (especially essential oil) is extracted with 10 times its wt. of CH_2Cl_2 and the CO compounds are converted into water-sol. deriv. by treating with *N*-dimethylglycine-hydrazide monochloride (Viscontini, *Helv. chim. Acta*, 1950, **33,** 1773). The extracted compounds are converted into their dinitrophenylhydrazones, and after purification applied to a radial chromatogram using paper impregnated with dimethylformamide as the stationary phase and a 5 : 3 mixture of cyclohexane–cyclohexene as the mobile phase. G. R. WHALLEY.

Preservatives

Use of sorbic acid in food. D. Florentin (*Ann. Falsif., Paris*, 1959, **52,** 454—459).—The fungicidal properties of sorbic acid and its uses in oenology are discussed. Its determination in wines by steam distillation followed by spectrophotometric assay is described. (13 references.) J. V. RUSSO.

Colouring matters

Isolation of caramel colour. Union Starch & Refining Co. Inc. (Inventors: J. E. Cleland and A. le R. Meyer) (B.P. 811,545, 12.3.57).—A caramelised carbohydrate solution, which contains substances inhibiting fermentation of sugars, is distilled (with addition of more water if necessary) to remove such substances and the remainder inoculated with an organism or enzyme which can ferment sugars present. Caramel colour bodies are present in the solution and can be isolated in relatively pure form. F. R. BASFORD.

Food Processing, Refrigeration

Thermal methods of food preservation. V. Kyzlink (*Industr. aliment. agric.*, 1959, **76,** 687—694).—The principles involved in the thermal destruction of microflora, the thermal inactivation of enzymes, and the retention of nutritive and organoleptic qualities in the process of commercial sterilisation of foods are discussed, and advances during the past decade in sterilisation technology and machinery are reviewed. (18 references.) C. L. HINTON.

Microbial spoilage of canned food. I. Isolation and identification of some spoilage bacteria. G. Rangaswami and R. Venkatesan (*Proc. Indian Acad. Sci.*, 1959, **50B,** 349—359).—*Bacillus circulans* Jordan, *B. brevis* Migula, *B. subtilis* Cohn, *B. coagulans* Hammer, *B. licheniformis* (Weighmann) Chester, *Lactobacillus fermenti* Beijerinck and *Clostridium histolyticum* (Weinberg and Seguin) Bergey *et al.* are shown to cause spoilage in cans of curried vegetable, sliced potato and mixed fruit. E. G. BRICKELL.

Heat resistance of *Bacillus subtilis* spores. I. Effects of chemicals. A. N. Bose and A. K. Roy (*J. sci. industr. Res.*, 1959, **18C,** 248—250).—The effect of NaCl, $CaCl_2$ sucrose, glucose and acetic, propionic and butyric acids on the resistance of *B. subtilis* spores to heat treatment during the canning of food was studied. Spores isolated from spoilt canned vegetables were suspended in M/15 phosphate buffer (pH 6·5). The spore suspension was added to the prepared sterile solutions in thermal death time tubes so that the concn. of spores in the solutions was 10^6/ml. The sealed tubes were kept at 110°. The total viable count was reduced to zero after 6 min.

heating with a 1% NaCl or 0·1% $CaCl_2$ solution. A 55% sucrose, 28·9% glucose or 0·05M acid solution reduced the heat resistance appreciably. The effect of the acid increased with increase in mol. wt. A very dil. NaCl solution enhanced the heat resistance of the spores. A. ABBOT.

Diffusion of sulphite during vegetable dehydration. R. B. Duckworth and M. Tobasnick (*J. Sci. Fd Agric.*, 1960, **11**, 226—228).—The distribution of sulphite in prepared samples of potato, carrot and cabbage, scalded in solutions containing labelled ^{35}S sulphite, and subsequently dehydrated and stored was studied. The sulphite penetrates the strip (e.g., of root vegetable) with slight concn. at the centre and in cabbage accumulates in the veins. Since sulphite is used to inhibit browning, it seems that sugars and amino-acids present in the sample diffuse similarly leading to formation of brown centres. E. M. J.

Dried edible products. R. A. S. Templeton (B.P. 810,218, 7.5.56).—A process for the prep. of dehydrated foodstuff (e.g., carrots) in the form of dice or small flakes, comprises admixing the foodstuff (in discrete pieces) with enough dried powdered food substance (e.g., cooked mashed potato powder) to absorb the surface moisture of the former without losing powdery consistency, then drying the pieces by evaporation (whilst mixed with or after separation from the powdered foodstuff). F. R. BASFORD.

Freezing of food. R. G. T. Baader (Inventor: S. Cooper) (B.P. 811,231, 10.7.56).—A device (figured and claimed) for the deep cooling and freezing of food comprises a mobile unit with several hollow shelves and means for varying the distance between adjacent shelves. F. R. BASFORD.

Frozen food product. J. F. Paulucci (B.P. 811,966, 2.5.57).—A method of preparing a frozen food package (especially Chow Mein) of improved quality comprises blanching Chinese food vegetable (while unseasoned) to inactivate enzymes (while maintaining crispness and taste of the vegetable); draining off surplus water; separately preparing a cooked sauce and seasoning it with enough seasoning for itself and for the vegetable; then placing the vegetable and the sauce in separate compartments of a single container; and freezing the packaged unit. F. R. BASFORD.

Perishable foodstuffs. Unilever Ltd. (B.P. 810,145, 24.8.56. Neth., 31.8.55).—A method for the manufacture of perishable foodstuffs (meat or fish), in the course of which the food is moved along a predetermined path and subjected to one or more processing operations, comprises forming a hard, smooth, plane surface by freezing a body of aq. liquid extending along the path, continuously feeding successive portions of foodstuff on to the frozen surface, and mechanically causing the portions to move progressively along the path while simultaneously supported and cooled thereon. Apparatus is figured and claimed. F. R. BASFORD.

Packaging

Errors caused by stainless steel cylinders in plate bioassays. N. S. Snell and J. C. Lewis (*Antibiotics & Chemotherapy*, 1959, **9**, 609—612).—Five out of seven antibiotics showed loss of potency after contact with stainless steel assay cylinders. With four of these, losses were greater with those that had become tarnished. It is suggested that this loss may be caused by adsorption but this aspect was not investigated. C. V.

I. Chemistry and properties of plastics. H. Hopff. **II. Plastics in modern packing industry.** K. Meyer. **III. Testing plastics for resistance to ethanol and spirits.** O. Wanger. **IV. Experience with plastics in dairy industry.** E. Flückiger. (*Mitt. Lebensm. Hyg., Bern*, 1959, **50**, 460—477, 478—492, 493—498, 499—507).—I. A review covering classification, methods of synthesis, and the relationship of properties to mol. composition and structure.

II. A review covering methods for shaping the appropriate types of plastics into packing-foils, containers, tubing, etc., and for the manufacture of protective varnishes.

III. The suitability of varnishes for coating storage tanks is often influenced by the method of application, the nature of the metallic surface to be coated, and (possibly) by variations in details connected with the manufacture of any particular batch. A resistance test is described in which the varnished test-plates or -rods are kept partly immersed in the methanolic samples contained in glass-stoppered cylinders; after 6 months (in darkness), the entire surfaces of the plates are examined for signs of breakdown in the film, and the smell and taste of the liquid samples are judged in comparison with reference samples. Most varnishes yield small amounts of higher alcohols (easily detected by the Komarowsky test) and fluorescent substances to methanolic liquids. A rapid sorting test for varnishes (provided that they are sufficiently heat-resisting) consists in suspending the test-plates during 24—48 hr. in the vapour from boiling (refluxing) 90—96% EtOH. The behaviour of the varnish under practical conditions cannot always be predicted by laboratory tests.

IV. The possible advantages and observed drawbacks of the use of polyethylene as material for milk-churns and -pails are considered in detail. Plastic churn-lids have, in practice, proved mechanically deficient and prone to abrasion on cleaning; the abraded surfaces are liable to fungal infection. Water-miscible polyvinyl acetate forms a very satisfactory film cover for finished export-cheeses; its use as a covering for ripening Emmental cheese reduces working costs by ~50% and diminishes losses by shrinkage without affecting the quality of the cheese. P. S. ARUP.

Packaged foodstuffs. T. Hedley & Co. Ltd. (B.P. 810,531, 14.1.57. U.S., 16.1.56).—There is claimed a packaged foodstuff prep. consisting of a mixture (**I**) of particulate baking ingredients suitable for making a shortening-containing cake (e.g., a mixture of flour, sugar and glyceride shortening agent), contained within a pliable bag of usable capacity not less than 157% of the bulk vol. of **I**, such that liquid can be introduced into the bag and incorporated into **I** to form a batter of improved properties. The bag is preferably made of polyethylene, copolymer of vinyl chloride or vinyl acetate, polyvinylidene chloride or rubber hydrochloride, of thickness 0·001—0·003 in. F. R. BASFORD.

Miscellaneous

Nutrition, proteins, amino-acids, vitamins

Effect of degree of polishing of rice on nitrogen and mineral metabolism in human subjects. G. Rama Rao, H. S. R. Desikachar and V. Subrahmanyan (*Cereal Chem.*, 1960, **37**, 71—78).—Experiments aiming at an optimal degree of polishing for rice which would not deplete thiamine below a safe level (1·5—1·8 μg. per g.) and which would not adversely affect mineral and N balances in human subjects are described. (19 references.) J. V. RUSSO.

Calcium fortification of rice. R. Radhakrishnamurthy, H. S. R. Desikachar and V. Subrahmanyan (*J. sci. industr. Res.*, 1959, **18C**, 245—247).—The possibility of increasing the Ca content of rice for human consumption was studied by (*a*) calcuring, i.e., soaking paddy or rice in $CaCl_2$ solutions during parboiling, (*b*) surface deposition by spraying a mixture of rice and $CaCO_3$ with a solution of acetic, citric or phosphoric acid, (*c*) spraying the rice with aq. Ca acetate, sometimes followed by a phosphate spray, (*d*) preparing a rice-shaped grain from $CaCO_3$ and flour which could be mixed with a large bulk of rice, 10 lb. paddy or 5 lb. rice samples of Ratnachudi type (S-749) being used for the tests. The efficiency of the Ca enrichment was judged by expressing Ca in the cooked rice as a % of that added. When a Ca acetate spray was used about 70% of the Ca remained after careful cooking; 80% of the original Ca was left in the cooked rice when a roasted premix of $CaCO_3$ (85%) and flour (15%) was added. A. ABBOT.

Action of different fats on frying of protein of beef. G. Varela, A. Pujol, O. Moreiras and C. Matéu (*An. Bromatologia*, 1959, **11**, 401—406).—Results of comparative tests by the Mitchell technique of the digestibility of beef protein after frying in olive oil, soya-bean oil, butter, margarine and lard, are reported. E. C. APLING.

Digestibility coefficient of various sausages. O. Moreiras, G. Varela and A. Pujol (*An. Bromatologia*, 1959, **11**, 381—393).—Results of determinations of digestibility, protein efficiency and palatability are reported for six different types of sausage, using white rats by the Mitchell technique. (17 references.) E. C. APLING.

Digestibility of various viscera. A. Pujol, G. Varela and O. Moreiras (*An. Bromatologia*, 1959, **11**, 395—400).—Results of determinations of digestibility and protein efficiency are reported for liver, heart, kidneys, tongue and brains from adult cattle, using white rats by the Mitchell technique. E. C. APLING.

New techniques in protein chemistry. E. T. Mertz (*Cereal Sci.*, 1960, **5**, 32—34, 38).—A new method of extracting protein from defatted maize germ and endosperm, using a reagent of high pH containing Cu and SO_3^{2-} and its application to different varieties of maize are described. The Cu extract contains 90—100% of the N depending on the variety of maize tested. (19 references.) J. V. RUSSO.

Nutritional effect of polymers isolated from thermally oxidised maize oil. E. G. Perkins and F. A. Kummerow (*J. Nutr.*, 1959, **68**, 101—108).—Weanling rats were fed 21 days on a diet of glucose (50), casein (31), Wesson salt (5), cottonseed oil (2) and test fat (12%),

water and fat-sol. vitamins. With the non-distillable residue from the non-urea-adduct-forming fatty acid of maize oil (**I**) death occurred in seven days; this fraction amounted to 30% of the original **I** and had been heated for 48 hr. at 200°. Using this diet with an equal vol. of fresh **I**, the animals survived the 21-day test period but growth deficiency caused by the non-urea-adduct-forming adduct of **I** was only partially counteracted. The major portion of the thermally oxidised **I** did not appear to be damaged by this severe treatment and animals fed on this (the urea-adduct-forming fraction) gained as much as those fed on fresh **I**. (12 references.)
C. V.

Determination of riboflavin and nicotinic acid in Italian wheats. M. Filajdić and M. Grüner (*Kem. u. Industr., Zagreb*, 1959, **8**, 299—300).—Riboflavin (**I**) and nicotinic acid (**II**) contents in 11 varieties of wheat of Italian origin were determined on grain samples harvested in 1958 in two different locations in Yugoslavia, **I** by the Snell and Strong, and **II** by the Snell and Wright microbiological assay methods. **I** contents were 0·126 ± 0·016 mg. in the samples from Maksidar estate and 0·124 ± 0·0059 mg. in those from Botinec. **II** contents were 4·54 ± 0·67 mg. and 3·81 ± 0·92 mg. per 100 g. of grain from the two estates respectively. The highest contents of **I** and **II** were in Fortunato (0·138 and 5·68), Abbodanca (0·147 and 5·12), and in R-16 (0·151 and 4·77) mg. per 100 g., respectively, wheat varieties. All determinations were made on the grain containing 12% moisture. A. L. GROCHOWSKI.

Human dietary preparations. Glaxo Laboratories Ltd. (Inventors: W. F. J. Cuthbertson and G. A. Childs) (B.P. 810,813, 11.11.55).—A solid human dietary prep., adapted for dispersion in aq. media (to provide a liquid food), comprises a pulverulent water-assimilable protein of low bulk *d* (4—6·5 c.c. per g.) and a stable assimilable bland fat, m.p. <25°, absorbed therein (~1·25 pt. per pt. of protein). The protein (in flake form) may be a caseinate or egg albumin, and the fat is preferably arachis oil, olive oil, sesame oil or cottonseed oil. If desired there may also be present additional protein (dried full cream or dried skim milk), sugar, vitamins, trace elements, etc. F. R. BASFORD.

Unclassified

Determination of hexachlorocyclohexane [BHC] residues in foodstuffs. R. Wood (*Analyst*, 1960, **85**, 21—24).—A hexane extract of the sample is treated with a mixture of conc. and fuming H_2SO_4 and after separation is passed through a Celite–H_2SO_4 column. The residue obtained by evaporation of the eluate at low temp. is boiled with malonic acid and acetic acid and then, when cold, is treated with more malonic acid, zinc dust and glacial acetic acid. The liquid is heated in the dechlorination apparatus of Hancock and Laws (*ibid.*, 1955, **80**, 665) with a sintered glass bubbler (described) immersed in a nitration mixture. The nitration mixture is extracted with methylene chloride and the washed and dried extract is evaporated and the residue is dissolved in ethyl methyl ketone. An aliquot is shaken with 50% KOH solution. and after development of the colour (*m*-dinitrobenzene) the extinction is measured and referred to a calibration graph. (12 references.)
A. O. JONES.

Effect of chemical environment on lethality of γ-radiation for anaerobic bacterial spores. N. J. Williams-Walls (*Dissert. Abstr.*, 1959, **20**, 1975—1976).—The sensitivity to γ-radiation of spores of *Clostridium botulinum* and *Clostridium* sp. (P.A. 3679) was not altered by changing the state of the medium from liquid to gel, or by change of pH in the range pH 3·2 to 8·4. Of 21 substances tested, nine protected the spores to some extent when present in the medium during irradiation; in order of increasing protective activity, these were: lysine, *p*-chloromercuribenzoate, $NaNO_2$, cysteine, gelatin, methionine, catalase, reduced glutathione and Na hydrosulphite. The protective effect of reduced glutathione was due to the SH end-group. Protection by Na hydrosulphite appeared to involve more than a simple removal of O from the medium.
M. D. ANDERSON.

Prevention of infections and alimentary toxic infections. The hygienic control of food industries and a bacteriological analysis of their products. D. A. A. Mossel, J. Bechet and R. Lambion (*Rev. Ferment.*, 1959, **14**, 187—205).—Bacteriological standards of quality for various types of food products are discussed. Techniques for the microbiological analysis of foodstuffs are enumerated and methods for the detection of fraudulent additions of antiseptics and antibiotics are discussed. J. V. RUSSO.

Behaviour of antibiotics in foods. II. W. Diemair and W. Rödder (*Z. LebensmittUntersuch.*, 1960, **111**, 365—371).—Aq. solutions of Aureomycin hydrochloride (**I**) heated in an autoclave at 121° and at 136°, and examined by the method previously described (cf. J.S.F.A. Abstr., 1960, i, 317) still retained some activity after 20 min. Most of the loss occurred during the respective preliminary heating-up periods of 4 min. and 5 min. In buffered solution and in milk, destruction was more rapid, but after 15 min, an addition of 120 μg./ml. to milk still retained 0·5% of its activity. Comparisons of the activity of reverin (**II**) (methyl pyrrolidinotetracycline) as citrate and **I** showed that ~10 times the amount of **II** was required as of **I** for the same inhibitory activity on *Bacillus subtilis*. With **II** in aq. solution at 4° and at room temp. a rise to about 160% of the initial activity occurred before inactivation began. This was caused by a splitting of **II** into the more active tetracycline and methylpyrrolidine. C. L. HINTON.

Release of iron from conjugates in foods. R. Sanford (*Nature, Lond.*, 1960, **185**, 533—534).—Further data on the extraction of Fe *in vitro* from foods emphasise the importance of cooking haemoglobin (blood) (**I**), and the variability with which Fe is released by acid peptic digestion. Thus, higher % are released from liver than from **I**, the Fe in the liver coming from tissue Fe rather than from contained blood. Sultanas and beet (**II**) yielded higher % than did **I**, but the abs. amounts were smaller. Phytic acid in **II** does not inhibit liberation of Fe. Revised dietary tables of Fe values are proposed. W. J. BAKER.

Determination of zinc with zincon, and protective action of polyphosphates [against loss] in determination [involving incineration] of trace elements. W. T. Binnerts (*Chem. Weekbl.*, 1960, **56**, 66—67).—Amounts of Zn ≮0·003% can readily be determined in biological material by wet combustion and the use of the zincon reagent. Small amounts of added Zn can be satisfactorily recovered without noteworthy interference by Cd, Co, Cu, Ni or Pb. The observation of Campen and Dumoulin that the presence of Ca polyphosphates prevents loss of Zn on incineration is paralleled by that of the author that loss of traces of I from milk ash at 600° is reduced by the presence of Ca^{2+} and phosphate. The addition of $CaHPO_4$ before incineration will probably conduce to the retention of trace elements. P. S. ARUP.

Food products. Unilever Ltd. (B.P. 810,762, 22.8.57. U.S., 23.8.56).—A protein food product in the form of a spread resembling meat is obtained by film-drying an aq. slurry containing oilseed (groundnut) protein and optionally an edible inner additive (starch, fat, flavouring material, salts and colouring matter); cutting the resulting sheets into small flakes (1—3 mm. wide, 0·05—0·2 mm. thick, pH 4·5—7 in aq. dispersion, ≮4 pt. of protein solids per pt. of water); and mixing the latter with an aq. emulsion of fat (cottonseed oil) optionally containing an edible outer additive (texturising and/or stabilising agents such as starch, fat, etc.); then hydrating the flakes. F. R. BASFORD.

Purification of *Bacillus subtilis* protease. Naaske & Co. Ltd. (Inventor: K. Okunuki, B. Hagihara and T. Ukita) (B.P. 810,566, 7.2.56).—A simple and an economical method for the recovery of *Bacillus subtilis* protease of high purity from solutions containing it comprises passing the solution through a coarse-porous cation-exchange resin (Kaken resin I, Duolite C-10, Duolite CS-101, Amberlite IRC-50, Duolite S-30 or Low Crosslinking Dowex-50) buffered to pH 6—8, then eluting the resin with a protease solvent of higher pH than the original solution, e.g., a buffer solution of pH 6·5—7·2. F. R. BASFORD.

Amino-acid compounds. Parke, Davis & Co. (B.P. 811,103, 24.4.56. U.S., 25.8.55 and 9.3.56).—Several methods of prep. are given for alkali or alkaline earth metal salts of 6-*diazo*-5-*oxo*-2-*aminohexanoic acid* or its low-mol. alkyl esters, which have phytotoxic and herbicidal properties. J.A.C. ABSTR.

Tenderising food. Reflectone Corp. (Inventor: L. G. Simjian) (B.P. 811,618, 16.9.57).—An apparatus for tenderising a normally deformable article of food (meat, grapefruit, oranges) which has been transformed from its deformable state to a substantially rigid state (by freezing) comprises one or more electromechanical means for transforming electrical energy into mechanical vibrations, and coupling means for transferring the vibrations to the article. Apparatus is figured. F. R. BASFORD.

3.—SANITATION

Insect pests of food and food handling. A. N. Johnston (*Food Tech. Aust.*, 1960, **12**, 19, 21, 23, 25, 26, 44).—Types of pests commonly encountered and methods of control are reviewed in relation to the producer down to the consumer. Conditions of location, development and speed of infestation refer in particular to grain beetles. Modern treatments include residual and space spraying with chlorinated hydrocarbons (e.g., DDT), pyrethrums and org. phosphates. P. M. KINGSTON.

Insect-fragment contamination in cereals. O'D. L. Kurtz and K. L. Harris (*Cereal Sci.*, 1960, **5**, 35—38).—Methods of recognising and identifying insect fragments in cereals are discussed using the granary weevil as an example. The text is illustrated with photomicrographs. J. V. Russo.

Determination of uric acid in wheat flour infested by *Tribolium castaneum* Duv., using paper chromatography. S. Venkatrao, K. Krishnamurthy, M. Swaminathan and V. Subrahmanyan (*Cereal Chem.*, 1960, **37**, 93—96).—Uric acid was determined in control and infested samples of flour by direct colorimetry and by paper chromatography and the results are compared. J. V. Russo.

Effect of infestation by *Tribolium castaneum* Duv. on quality of wheat flour. S. Venkatrao, R. N. Nuggehalli, S. V. Pingale, M. Swaminathan and V. Subrahmanyan (*Cereal Chem.*, 1960, **37**, 97—103).—The effect of insect infestation on a hard wheat flour was assessed by means of chemical and baking tests. (12 references). J. V. Russo.

Toxicity to house-fly larvae of insecticides administered as single oral doses to chicks. M. Sherman and E. Ross (*J. econ. Ent.*, 1959, **52**, 719—723).—The insecticides were given as capsules or introduced directly into the crop. One week after the treatment the LD_{50} in mg./kg. body wt. was diazinon, 8·4; Dipterex, 65; malathion, 37; Dow ET-14, 890 and Dow ET-15, 1180. Phenothiazine was non-toxic. Except for diazinon and phenothiazine all reduced the wt. gain of survivors. The toxicity of chick faeces to house-fly larvae was directly related to dosage and inversely to time after ingestion. More than 90% mortality occurred for 1 day with malathion, 2 days with Dipterex, 3 days with Dow ET-14, and 6 days with Dow ET-15. (14 references.) C. M. Hardwick.

Cross resistance in a diazinon-resistant strain of *Musa domestica* (L.). A. J. Forgash and E. J. Hansens (*J. econ. Ent.*, 1959, **52**, 733—739).—Diazinon (**I**)-resistant house flies were exposed to **I** for nine generations which then showed increased resistance to **I** and to 18 other insecticides; for chlorinated hydrocarbons this was 1000 to 6000-, chlorinated cycloalkanes 200 to 400-, phosphorus compounds 5 to 38-, carbamates 10 to 37- and Dilan 10-fold. Pyrethrins, allethrin and Lethane had lower values which may not be due to true resistance. Tests on field collected flies gave similar results. (35 references.) C. M. Hardwick.

Reproduction following insecticidal treatment in two resistant strains of house flies. P. E. Hunter, L. K. Cutkomp and A. M. Kolkaila (*J. econ. Ent.*, 1959, **52**, 765—766).—Females from two DDT-resistant strains were treated with DDT and diazinon. All treated flies had a shortened life span and laid fewer eggs per clutch. Diazinon-treated flies from one strain showed a higher egg fertility than the controls. DDT-treated flies from both strains laid fewer eggs than the controls or the diazinon-treated batch. C. M. Hardwick.

Dilan formulations for adult house-fly control. H. F. Schoof and J. W. Kilpatrick (*J. econ. Ent.*, 1959, **52**, 776—777).—Dilan + DDT + cottonseed oil and Dilan + cottonseed oil sprays failed to give residual control in dairies in May comparable with that obtained the previous autumn. C. M. Hardwick.

Effectiveness of Ronnel as a cord impregnant for house-fly control. J. W. Kilpatrick and H. F. Schoof (*J. econ. Ent.*, 1959, **52**, 779—780).—In six dairies satisfactory control lasted 8—16 weeks. In one dairy where control was poor, parathion-impregnated cords also failed. C. M. Hardwick.

Detection and estimation of the biologically active constituents of pyrethrum. H. J. Smith (*J. Sci. Fd Agric.*, 1960, **11**, 172—176).—This method for determination of pyrethrin I, cinerin I, pyrethrin II, cinerin II, by quant. prep. and chromatographic separation of the 2,4-dinitrophenylhydrazones is an extension of that described, cf. J.S.F.A. Abstr., 1959, ii, 104; all four constituents may be quant. separated on Al_2O_3 columns. The method is rapid and suitable for routine analysis. E. M. J.

Comparative residual behaviour of pyrethrins and piperonyl butoxide on wheat. R. C. Blinn, R. W. Dorner and F. A. Gunther (*J. econ. Ent.*, 1959, **52**, 703—704).—Analysis by chromatographic and colorimetric methods showed pyrethrins to have a half-life of 5·8 weeks and piperonyl butoxide 9·9 weeks. This should be borne in mind when analysing residues by ascertaining the quantity of piperonyl butoxide present. C. M. Hardwick.

Synergistic action of piperonyl butoxide with Bayer 21/199 and its corresponding phosphate in mice. W. E. Robbins, T. L. Hopkins and D. I. Darrow (*J. econ. Ent.*, 1959, **52**, 660—663).—The toxicities of Bayer 21/199 [*O*-(3-chloro-4-methylumbelliferone) *OO*-diethyl phosphorothioate] and its phosphate were increased 4—6-fold when piperonyl butoxide was administered by the same or a different route. *In vitro* it had no effect. Piperonyl butoxide prevented the enzymic hydrolysis of labelled Bayer 21/199 to more polar products. Radioactive products could be detected only in the liver. C. M. Hardwick.

Estimation of total phosphate in water. R. S. Robertson (*J. Amer. Wks Ass.*, 1960, **52**, 483—491).—The normal test wherein the complex condensed phosphate is changed into the orthophosphate followed by Mo-colorimetric methods is discussed; the results obtained are often low and the complete degradation may require 1—4 hr. The test described can be carried out in ~5 min. heating time and is specially designed for field use. It is intended for the determination of all sol. inorg. phosphates and phosphate complexes in water but it has also been used with adenosinetriphosphate successfully. Normal methods partially destroy the org. part of the mol. (10 references.) C. V.

OO-Dialkylphosphoric or OO-dialkylthionophosphoric acid esters of N-(hydroxyalkyl)lactams. Badische Anilin- & Soda-Fabrik A.-G. (B.P. 810,646, 31.5.57. Ger., 5.6.56).—The compounds, useful as pesticides (especially active against *Musca domestica* and aphids), are obtained by interaction of $OR(OR')\cdot PX\cdot Y$ with $[-[CH_2]_m\cdot CX\cdot N\cdot C[R''R''']_n\cdot OH$ at 10—100° in anhyd. solvent medium in presence of acid-binding agent [R and R' are alkyl of 1—4 C; X is O or S; Y is halogen; R'' and R''' are H, alkyl of 1—4 C optionally substituted by halogen (preferably Cl), alkenyl, aryl, or cycloalkyl; *m* is 3—7; *n* is 1—4]. The prep. of (2-*oxopyrrolidino*)-*methyl thionophosphate* by conventional means is described. F. R. Basford.

Emulsification of water-insoluble organic solvents. General Aniline & Film Corp. (B.P. 809,657, 21.11.55. U.S., 17.12.54).—A composition, for use in emulsifying a water-immiscible org. solvent (especially solvent employed in the prep. of insecticidal concentrates), consists of a water-sol. synthetic non-ionic surface-active agent containing at least 2 alkenoxy groups and derived from an alkyl(4—24 C)phenol(50—90) and the cyclohexylammonium salt (10—50 wt.-%) of an alkyl (<3 C)-benzenesulphonic acid, e.g., cyclohexylammonium dodecylbenzenesulphonate. F. R. Basford.

Bacterial digestion of organic matter. Dorr-Oliver Inc. (B.P. 801,144, 1.7.55. U.S., 1.7.54).—In the anaerobic digestion of bacterially digestible org. sludge (sewage sludge or industrial waste, e.g., from fermentation processes or packing house operations), pH is controlled by treating the sludge with carrier gas of sufficiently low CO_2 content such that it is able to absorb dissolved CO_2 from the sludge. The treatment is specifically effected with stirring in a confined zone in communication with the digesting sludge mass. Flow diagram is figured. F. R. Basford.

4.—APPARATUS AND UNCLASSIFIED

Apparatus for determination of water by distillation method L. Hübschen (*Z. LebensmittUntersuch.*, 1960, **111**, 371—373).—In the apparatus the mixed vapours of water and immiscible solvent emerge into the condenser at its top end, and in condensing wash down the water completely from the sides of the tube into the measuring capillary. Complete recoveries of known amounts of water up to 4 ml. were obtained after 20 min. The apparatus is particularly suitable for tobacco, 20 g. with 300 ml. xylol being distilled for 60 min. C. L. Hinton.

Bibliography of the biological effects of thorium. E. Hutchinson (*U.S. at. Energy Comm.*, 1960, Rep. UR 563, 47 pp.).—A survey covering, e.g., effects on vegetation, development of bacteria in soil, plant growth, yeast and yeast hydrogenase, and medical aspects (530 references.) C. V.

A bibliography of biological applications of autoradiography, I. 1954/57. II. 1958/59. M. E. Johnston (*U.S. at. Energy Comm.*, 1958, 1959, Rep. UCRL 8400, 8901; 41, 22 pp.).—I. (536 references.) II. (312 references.) C. V.

Uptake and distribution of strontium in vegetables and cereals. R. B. Duckworth and J. Hawthorn (*J. Sci. Fd Agric.*, 1960, **11**, 218—225).—Strontium-89 was applied continuously throughout the growth period to roots of species of cabbage, root vegetables and cereals grown in sand. The general distribution of absorbed Sr is discussed and auto-radiographs from mature plants are given. Sr may accumulate in parts of food plants not included in human diet but the discarded activity in parts fed to animals will lead back along the food chains to man. From data on field crops higher $^{89}Sr/^{90}Sr$ ratios are reported for outer leaves of cabbage as compared with inner leaves. ^{90}Sr/stable Sr ratios for bran are higher than those for flour. (22 references.) E. M. J.

Journal of Applied Chemistry

The following papers are appearing in the July, 1960, issue

Errors in infra-red absorption analysis due to solute–solvent interactions. II
By W. R. Ward

New Nessler reagent and its use in the direct nesslerisation of Kjeldahl digests
By K. R. Middleton

Solvent extraction of mineral acids. IV. Solute-solvent interaction in the system perchloric acid–water–tri-n-butyl phosphate
By A. S. Kertes and V. Kertes

Catalytic hydrogenation of sugars
By C. Boelhouwer, D. Korf and H. I. Waterman

Some viscous and elastic properties of rubberised bitumens
By L. M. Smith

Weathering of road tar in open-textured surfacings
By E. D. Tingle and N. Wright

Inductive effects and rate and equilibrium constants of chemical reactions
By J. C. McGowan

Tomato-red

Nestling at the foot of the eternally snow-capped Andes is lovely, flower-strewn Mendoza. Here is a happy place; a sunny, fertile garden in which vines, tomatoes and other crops thrive . . . if they are allowed to. The 'serpent' in this 'garden' is the root-knot nematode—a tiny, worm-like creature that lives on the roots of many crops throughout the world. In Mendoza, tomato plants have wilted and died in the sun, yields have declined . . . part of the havoc wreaked by the unseen nematodes.

Shell D-D is the prescription—and the remedy. In nematode-infested fields, the authorities injected this yield-increasing soil fumigant two weeks before planting. At harvest time, the tomatoes were picked and weighed. The crop was *three and a half times greater* than that from untreated ground.

Hope had become reality. With D-D, land that had become unfi for tomato-growing was brought into cultivation again. Today the fields around Mendoza blush red with tomatoes. Similarly with other crops throughout the world, D-D has rid the soil o nematodes and brought new hope—and new wealth—to grower everywhere. Between them, D-D, Nemagon, aldrin, dieldrin endrin and Phosdrin can control virtually every major world pest

D-D

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CONTENTS