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UTILISATION OF DIETARY ENERGY BY POULTRY. II.*—Effects of Indigestible Organic Matter and Protein on the Utilisation of Metabolisable Energy for Growth

By J. DAVIDSON, I. McDONALD, J. MATHIESON and R. B. WILLIAMS

Rations compounded from individual cereals or from mixtures of cereals were fed to growing cockerels. When the levels of protein and metabolisable energy were closely controlled, the effect of indigestible material up to levels likely to be encountered in practice did not appear to have a consistent effect on the utilisation of metabolisable energy for gain in tissue energy, although a maize plus wheat diet which had the lowest content of indigestible material proved superior to other rations tested.

A ration based on oats which contained the highest level of indigestible matter, gave poor growth, although utilisation of metabolisable energy for gain was normal. The poorer growth was attributed to reduced food intake owing to its relative unpalatability and bulk.

When fed a diet containing a critical content of protein with an amino-acid imbalance, the birds tended to compensate by consuming more food and thus more energy than was normal for their weight and the excess energy was lost as heat.

Introduction

Dietary metabolisable energy (ME) is transformed in the body either to heat energy or tissue energy in proportions depending on the balance of nutrients in the diet. Thus it is important that the balance of nutrients is such as to promote a maximum production of tissue energy. In the experiments to be described, a study has been made of some dietary factors affecting the utilisation of ME for tissue gain in chicks up to 4 weeks old and in young table cockerels up to 12 weeks old. The degree of utilisation was measured by determining the amounts of energy, protein and fat laid down in the birds.

Experimental (Expts. 1–3)

Analytical methods

Body analysis.—Chicks were prepared for analysis as follows: the alimentary tract was removed from the bird and cleaned of food and faecal residues; surplus water was removed with filter paper and the tract returned to the bird. After being weighed, the ' empty ' bird was put through a household meat mincer and the weighed mince dried in a freeze-dryer to a sponge-like cake containing about 2% of water. This material was weighed, sampled for determination of dry matter and then minced again. The resultant material was stored in screw-cap bottles with waxed liners until required for analysis.

Older birds were dealt with in a similar way but it was necessary to take only a portion of the minced material for freeze drying.

Protein was determined by the Kjeldahl method with mercuric oxide catalyst.

Fat was estimated by a ' warm extraction ' procedure, the Soxhlet thimble being suspended above the boiling diethyl ether.

Calorific value was determined by bomb calorimetry.

Diet analysis.—Proximate analysis and the determination of pentosans and cellulose were carried out by the methods of the A.O.A.C.,[1] starch and sugars by the anthrone method of Clegg,[2] and prediction of the metabolisable energy content of feeds by that of Carpenter & Clegg.[3] Indigestible organic matter (IOM) was taken as 10% of the sum of crude protein plus crude fat, 96% of the pentosans[4] and 100% of the cellulose and unaccounted material some of which would be lignin.

Effect of feeding equal amounts of metabolisable energy from predominantly maize and predominantly barley diets (Expt. 1)

Diets

In this experiment two diets were fed which differed in their ratios of crude protein to energy.

* Part I: *J. Sci. Fd Agric.*, 1957, **8**, 173

This was simply achieved by feeding the same supplementary protein and vitamin mixture with either maize which contains about 3·3 kcal. ME per g., or barley with about 2·8 kcal. ME per g. The diets, shown in Table I along with those for Expt. 2, differed also in their content of IOM.

Table I

Composition of diets in Expts. 1 and 2

Experiment	1		2		
Diet no.	312 (Maize)	311 (Barley)	469 (Maize)	467 (Barley)	468 (Barley)
Barley, ground	—	77	—	68	51
Maize, ground	77	—	77	—	—
Maize starch	—	—	—	18·5	25
Casein	—	—	—	—	2
Arachis oil	—	—	—	—	2
Supplement mix	23	23	23	23	23
Totals	100	100	100	109·5	103
Proportions fed	87·2	100	100	109·5	103
ME content, kcal./kg.	3130	2730	3040	2860	3080
IOM content, %	12·5	17·2	13·0	15·4	13·6
Protein/ME, g./kcal.	0·058	0·073	0·058	0·059	0·058
IOM/ME, g./kcal.	0·040	0·063	0·043	0·054	0·044

Supplement mix	
White fish meal	13·5
Grass meal	6·0
Casein + vitamins* + manganese†	3·0
Cod liver oil	0·5
	23·0

* Sufficient to supplement final diet with riboflavin 0·5 mg./kg. (Expt. 1) or 1 mg./kg. (Expt. 2); and calcium pantothenate 1 mg./kg. (Expt. 1) or 2 mg./kg. (Expt. 2)

† Expt. 2 only, sufficient to supplement final diet with manganese as sulphate 15 mg./kg.

Method

The procedures were similar to those described in a previous paper.[5] Sixty Light Sussex cockerels were chosen from 200 at 1 week old; 6 were killed to provide the data on initial gross energy value, 48 were randomised to treatments and cages, and the remaining 6 were kept for a time as spares, 3 on each treatment. Only one of these was required, to replace a chick which died.

The birds on the barley diet received daily an amount of food equivalent to 16% of their average live body weight as measured every second day; those on the maize diet received a smaller ration, providing an equal amount of ME, the ME values having been measured in a preliminary experiment with young chicks. The feeding levels used were just below *ad libitum* feeding and ensured that all food was eaten. Food spilled into droppings trays and water pots was weighed and taken into consideration in the final calculations.

After 3 weeks each experimental bird was killed and treated as previously described. The protein, fat and gross energy contents of each bird were then determined.

Results

The utilisation of metabolisable energy for gain shown in Table II indicates that the mean gains in total energy, fat and body weight were all significantly higher on the high-energy maize diet than on the low-energy barley diet. The total energy gain was about 15% higher on the maize diet.

From Table III, which shows the relative amounts of energy and nutrients consumed, it can be seen (i) that differences in intake were considerable for protein and IOM and (ii) that the intake of readily available energy in the form of starch and sugars was approximately the same in both diets.

The more obvious factors which may have given rise to the lower utilisation of ME on the barley diet were the greater protein intake, an amino-acid imbalance in the protein or the larger amounts of energy required for gizzard activity and moving the more fibrous barley diet in the alimentary tract.

Table II

Utilisation of metabolisable energy for productive gain in Expt. 1

	Maize diet 312	Barley diet 311	Standard error of difference
ME eaten per bird, kcal.	955	954	±2
Energy gain per bird, kcal.	181	157	±4·8
Fat gain per bird, g.	4·9	2·8	±0·3
Protein gain per bird, g.	24·1	22·9	±0·6
Live weight gain per bird, g.	124	117	±3

Table III

Amounts of energy and nutrients consumed per bird between 7 and 28 days of age in Expt. 1

	Maize diet 312	Barley diet 311
ME, kcal.	955	954
Total dry matter, g.	263	300
Crude protein, g.	55	69
Crude fat, g.	13	11
Ash, g.	13	17
Starch, g.	144	142
Sugars, g.	7	8
Pentosans, g.	15	25
Cellulose, g.	10	16
Unaccounted, g.	6	12
IOM, g.	37	60

Fig. 1 shows the amino-acid composition of each diet corresponding to 300 kcal. ME, compared with amino-acid requirements for chicks published by the National Research Council (NRC) of the United States, 300 kcal. ME being a value assumed to be near the energy content of 100 g. of the rations for which the NRC give their requirement figures.[6] The amino-acid composition of each diet was estimated from a table of averages compiled by de Man & Zwiep.[7] From Fig. 1 it can be seen that divergence from the amino-acid requirements proposed by the NRC was greater for the barley diet and that leucine was probably the only amino-acid consumed in greater quantity by birds on the maize diet.

Effect of feeding equal amounts of metabolisable energy from predominantly maize and predominantly barley diets when the intakes of protein and indigestible organic matter are approximately the same (Expt. 2)

Diets

These are shown in Table I. The formula for maize diet 469 was the same as for diet 312 fed in Expt. 1—namely 23 parts of a supplement mix and 77 parts of ground maize. Barley diet 467 was made up by adding to 23 parts of supplement mix, sufficient barley and maize

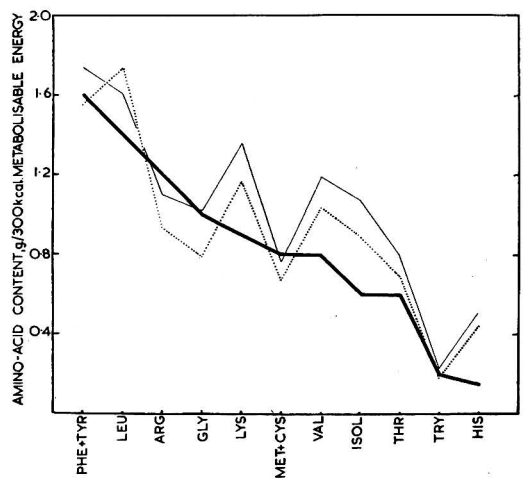

FIG. 1.—*Predicted amino-acid composition of diets in Expt. 1*

starch to contribute approximately the same amount of crude protein and ME as the 77 parts of maize. This diet contained about 25% more IOM per unit of ME than the maize diet. Barley diet 468 was made up by adding to 23 parts of supplement mix, sufficient barley, casein, arachis oil and starch to contribute approximately the same IOM, protein, ether extract and ME, respectively, as that provided by 77 parts of maize. This diet contained approximately the same amounts of protein and IOM per unit of ME as the maize diet.

The true ME of each diet was determined biologically during the experiment from calorific values of the food and corresponding droppings.

Method

Two hundred day-old Brown Leghorn × Light Sussex cockerels were fed a mixture of equal parts coarsely ground maize and coarsely ground wheat for 3 days and then the barley diet 467 for 4 days. At one week old they were weighed, extremes in weight discarded, and 66 randomised to treatments and cages—16 plus 2 spares plus 4 controls being allocated to each of the three treatments. The birds on diet 469 received a weight of food equal to 14% of their average live body weight each day and those on diets 467 and 468 sufficient to provide an equal amount of ME as calculated from predicted ME values. On experimental day 4 the control birds were killed for analysis as previously described and the unwanted spares were discarded. On experimental day 25 all remaining birds were killed for body analysis. Spillage was fed back.

Results

Table IV shows that the ME values predicted from the chemical analysis were, on the average, only 1% higher than those determined biologically during the experiment. The biological values are used in the calculation of energy uptake and utilisation.

Table V shows the average amounts of ME eaten by the birds in each group, the energy gains before and after adjustment to identical ME intake, the fat, protein and live weight gains and the percentages of protein retained. The differences between average figures were not significant. Table VI shows the amounts of nutrients consumed corrected to an equal energy intake.

Table IV

Metabolisable energy predicted and found in Expt. 2

	Predicted from equation,* kcal./kg.	Found by biological trial, kcal./kg.
Maize diet 469	3070	3040
Barley diet 467	2910	2860
Barley diet 468	3090	3080

* ME in kcal./kg. = 0·59(% dry matter) + 38[% protein + 2·25(% ether extract) + 1·1(% starch) + % sugars]

Table VI

Relative amounts of nutrients consumed per bird between 11 and 32 days of age in Expt. 2 when intake of ME is 1275 kcal.

	Maize diet 469	Barley diet 467	Barley diet 468
Total dry matter, g.	357	385	360
Crude protein, g.	74·5	75·9	74·5
Ether extract, g.	15·1	7·2	14·5
Ash, g.	17·2	19·6	18·2
Starch, g.	195·6	212·0	196·0
Sugars, g.	8·9	9·8	7·7
Pentosans, g.	20·7	25·6	19·2
Cellulose, g.	13·2	16·8	14·0
Unaccounted, g.	11·8	18·1	15·9
IOM, g.	53·9	67·8	57·2

Table V

Utilisation of metabolisable energy for productive gain in Expt. 2

	Maize diet 469	Barley diet 467	Barley diet 468	Standard error of difference
ME eaten per bird, kcal.	1256	1283	1288	±6
Energy gain per bird, kcal.	282	280	290	±8
Energy gain per bird adjusted to 1275 kcal. ME intake	290	277	285	±9 approx.
Fat gain per bird, g.	9·7	8·8	10·2	±0·9
Protein gain per bird, g.	34·6	35·2	34·6	±0·8
Live weight gain per bird, g.	179	182	177	±4·4
Protein retention, %	47·1	46·2	46·0	±1·1

No significant differences found

Fig. 2 which is again based on figures computed from tables by de Man & Zwiep shows that the relative amounts of the constituent amino-acids fed in diets 467, 468 and 469 were similar, apart from the leucine content of the maize diet which was again higher than that of the barley diets.

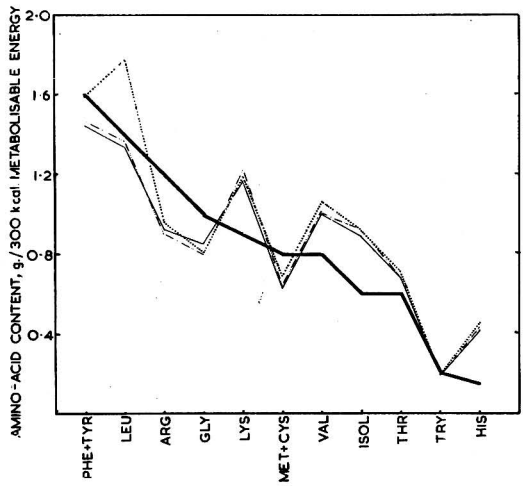

FIG. 2.—*Predicted amino-acid composition of diets in Expt. 2*

Effect of feeding equal amounts of metabolisable energy from rations based on barley or mixtures of wheat and oats when the intakes of protein and indigestible organic matter are the same (Expt. 3)

Diets

Table VII gives the formulae for the three diets fed. They contained the supplement mixture of white fish meal, grass meal, casein and vitamins used in Expts. 1 and 2. The cereal

Table VII

Composition of diets for Expt. 3

	Barley diet 607	Wheat/oats diet 608	Oats/wheat diet 609
Barley, ground	77·0	—	—
Wheat, coarsely ground	—	40·0	20·0
Oats, Sussex ground	—	20·0	37·0
Oat feed	—	5·0	—
Casein	—	0·5	1·5
Maize starch	—	9·5	17·5
Supplement mix	23·0	23·0	23·0
Proportions fed	100·0	98·0	99·0
ME content, kcal./kg.	2600	2650	2630
IOM content, %	20·6	21·1	20·9
Protein/ME, g./kcal.	0·074	0·073	0·073
IOM/ME, g./kcal.	0·079	0·079	0·079

Supplement mix	
White fish meal	13·5
Grass meal	6·0
Casein + vitamins* + manganese*	3·0
Adisco†	0·5
	23·0

* Sufficient to supplement the final diet with riboflavin 1 mg./kg., calcium pantothenate 2 mg./kg. and manganese as sulphate 15 mg./kg.

† Proprietary compound containing 1000 i.u. of vitamin A and 200 i.u. of vitamin D per g.

protein and energy were derived from barley, from 2 parts wheat plus 1 part oats, or from 2 parts oats plus 1 part wheat. The diets did not differ appreciably in content of leucine or any other important amino-acid (see Fig. 3). Small differences in the content of protein and IOM were adjusted by the addition of casein or oat feed. The protein/ME ratio was made the same in each diet by the addition of maize starch. ME values were again predicted by the method already cited[3] and used in calculating the amounts of each diet which were equivalent in metabolisable energy. Reference was made to those equivalents during the experiment when changing dietary allowances.

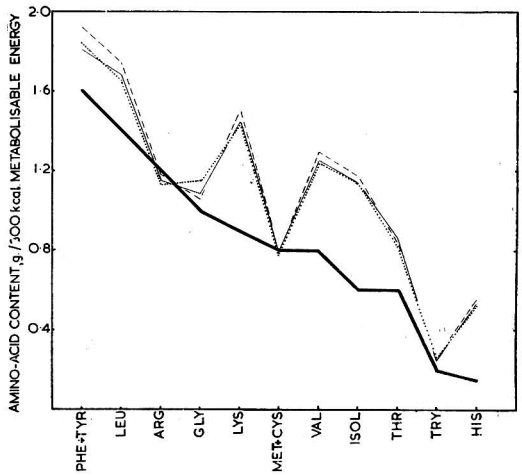

FIG. 3.—*Predicted amino-acid composition of diets in Expt.* 3

Method

Day-old White Leghorn × Rhode Island Red cockerels (150) were fed a mixture of equal parts coarsely ground maize and coarsely ground wheat for 3 days and then the barley diet 607 for 4 days. They were then weighed, extremes in weight discarded, and 48 plus 6 spares plus 12 for slaughter randomised to individual cages and diets. As in Expt. 1, each chick on the barley diet received daily a ration equivalent to 16% of the average live body weight of the group, a level just below *ad libitum* feeding. Birds on the other two diets received a ration sufficient to provide an equal amount of ME. After 4 days in which those birds which did not settle down to the experimental regime were discarded, the 12 control birds were killed for carcass analysis and unwanted spares were discarded. On experimental day 25, all remaining birds were killed for carcass analysis. Food spillage was fed back and droppings were collected every second day during the whole experimental period so that the true ME values could be determined and used in the final calculations of energy utilisation.

Results

The ME values predicted for each diet after chemical analysis are compared in Table VIII with the true values found during the experiment. The agreement to within 2% is good.

The live weight, protein and fat gains shown in Table IX have been adjusted for small differences in the true ME intakes. There was no difference in utilisation of the ME for energy gain from the equal amounts of ME, protein and IOM fed. This would indicate that the source of cereal protein and energy in such diets where protein is not limiting is without effect on growth.

Table VIII

Metabolisable energy values predicted and found in Expt. 3

	Predicted from equation, kcal./kg.	Found by biological trial, kcal./kg.
Barley diet 607	2650	2600
Wheat/oats diet 608	2690	2650
Oats/wheat diet 609	2660	2630

Table IX

Live weight, protein and fat gains per bird between 11 *and* 32 *days of age from equal intakes of metabolisable energy in Expt.* 3

	Barley diet 607	Wheat/oats diet 608	Oats/wheat diet 609	Approx. standard error of difference
Live weight gain per bird, g.	170	171	170	±5
Protein gain per bird, g.	33·5	34·5	34·0	±1·1
Fat gain per bird, g.	5·4	6·3	5·3	±0·6

Summary and conclusions from Expts. 1–3

When a high-energy diet based on maize was compared with a lower-energy diet based on barley, the resultant utilisation of ME for gain in young cockerels up to 4 weeks old was about 15% less on the latter, which contained 5% more IOM and had a higher ratio of protein to ME.

When both the IOM content and the protein to ME ratio in the barley diet were decreased to those of the maize diet by adjustment with starch and casein, the difference in utilisation of ME was only about 2% and was not statistically significant.

When the protein/ME ratio in the barley diet was reduced to that of the maize diet by adding starch only, and the IOM content was left about 2½% higher than that of the maize diet, the utilisation of ME was 4% less than on the maize diet and the difference was again not significant.

When the IOM content and the protein/ME ratios of diets based on mixtures of wheat plus oats were adjusted to that of a diet based on barley by the use of starch, casein and oat feed, the utilisation of ME was the same as that of the barley diet.

If the 15% difference in energy utilisation in Expt. 1 is not wholly due to the difference in quality and quantity of protein, it could have arisen from the increased IOM of the barley ration or to a combination of these factors. That still other factors may be involved is quite possible but it can be seen that when protein and IOM intakes are more closely related to energy intakes as in Expts. 2 and 3, the differences in energy utilisation tend to disappear. How well the effects due to varying the quantity and quality of protein can be distinguished from those due to changing the amount of IOM, is a matter for speculation as it is difficult to alter the IOM and protein contents of the rations without at the same time altering the amino-acid balance.

Birds may become less sensitive to changes in amino-acid balance and IOM as they grow older and to test the overall effect of differences in protein, energy and IOM on cockerels up to the age of table birds, the following broiler-type experiments were set up. They involved *ad libitum* feeding of groups of 12 birds up to 12 weeks to produce table birds of 3–4 lb. live weight.

Experiments 4 and 5

Effect of widely different percentages of dietary indigestible organic matter on the utilisation of dietary energy by cockerels during their first 12 *weeks of life* (Expt. 4)

Diets

These were based on wheat plus maize, barley or oats in order to provide diets varying considerably in IOM content. The starting diets are shown with certain analytical values in Table X. Diet A with wheat plus maize and diet C with oats were of low and relatively high IOM content, respectively, and represented the extremes likely to be encountered in practice.

At 6 weeks of age the percentage of protein in each diet was dropped by about 2% by substituting cereal for 2% white fish meal and 2% groundnut meal.

Table X

Composition of starting diets for Expt. 4

	Wheat and maize starting diet A	Barley starting diet B	Oats starting diet C
Wheat, coarsely ground	35·0	—	—
Maize, ground	35·0	—	—
Barley, ground	—	70·0	—
Oats, Sussex ground	—	—	70·0
White fish meal	13·5	13·5	13·5
Groundnut meal	7·0	7·0	7·0
Grass meal	3·0	3·0	3·0
Arachis oil	—	1·0	—
Maize starch	—	3·0	10·0
Adisco + B_2[a]	0·5	0·5	0·5
Dried yeast	1·0	1·0	1·0
Common salt	0·5	0·5	0·5
Ground limestone	1·0	1·0	1·0
Manganese sulphate	0·013	0·013	0·013
Aurofac 2A[b]	0·15	0·15	0·15
	96·7	100·7	106·7
ME predicted from chemical analysis, kcal./kg.	2840	2740	2620
Crude protein, %	20·7	19·8	19·0
IOM (calc.), %	12	18	24
Protein/ME, g./kcal.	0·072	0·072	0·073
IOM/ME, g./kcal.	0·042	0·066	0·092

[a] Proprietary mixture containing vitamin A 1000 i.u., vitamin D 200 i.u. and riboflavin 500 μg. per g.
[b] Proprietary mixture containing Aureomycin 3·6 g. per lb.

Method

Day-old Brown Leghorn × Light Sussex cockerels (500) were fed on equal parts of coarsely ground maize and wheat for 4 days and then weighed, extremes in weight being discarded. The chicks were randomised to 15 experimental groups of 12 birds and 1 spare group of 12 birds. The 16 groups were housed within the same hut in separate pens, each with an electrically heated hover, and wood shavings on the floor. Five experimental groups were allocated at random to each of the three diets. The spare group was fed diet B and used as a source of replacements for the experimental groups within the first two experimental weeks. The spare group was then discarded.

Previous experience had shown that food spillage into the water pots was negligible, but that round the feeding trough could on occasion be as much as 10% of the food eaten, especially during the first 3 weeks. To allow for this the feeders were placed on half-inch mesh wire netting stretched over large metal trays. Twice or thrice each week, as occasion demanded, the spilled food in each tray was separated from the wood shavings and droppings by a sieving technique and weighed. Separated spillage samples were analysed periodically for moisture and protein to ensure that the weights recorded represented spilled food. Between 6 and 12 weeks of age a specially designed feeding trough reduced spillage to less than 1%.

Bird weights and food eaten were recorded weekly. At 12 weeks of age six birds from each group were chosen for carcass analysis by arranging the group in weight order and choosing at random one for killing from each pair in ascending weight order. The intestinal tract contents were removed as for smaller birds and the birds minced as a group. Samples from the thoroughly mixed mince were freeze-dried and then ground prior to analysis for protein, fat and gross energy.

The carcass analysis of the chicks at the start of the experiment was predicted from results obtained in previous experiments. The error likely to arise from this prediction was negligible for the purpose of calculating the energy, fat and protein gains over the experimental period of 12 weeks.

Results and discussion of Expt. 4

The average spillage figures (2·3, 2·8 and 1·3% respectively in the diets A, B and C over the whole 12 weeks) were taken into consideration in the final calculations. It will be seen from Table XI that there was little difference in the final live weights of the birds on diets A and B but a significant drop in the weights of birds on diet C, the oats diet. The intake of food had been expected to increase from diets A to B to C because of the decrease in dietary metabolisable energy from 285 to 274 to 262 kcal./100 g. There was no simple correlation, however, between food eaten and ME contents of the diets.

Table XI

Live weights at 12 weeks and the amounts of food and certain components consumed in Expt. 4

	Wheat + maize diet A	Barley diet B	Oats diet C	Standard error of difference
Mean final weight per bird, g.	1854	1821	1720	±37
Mean food eaten per bird, g.	5490	5920	5750	
Food conversion ratio, g. food/g. gain	3·04	3·34	3·45	±0·03
Protein consumed per bird, g.	1030	1100	990	
IOM consumed per bird, g.	660	1070	1380	
Utilisation of ME for gain, %	23·0	20·6	20·2	±0·6

There were dietary differences in the food conversion ratios, which were significantly better on maize plus wheat diet A than on barley diet B or oats diet C ($P < 0{\cdot}001$). The conversion ratio was better on barley diet B than on oats diet C ($P < 0{\cdot}01$). These differences confirm the well-known fact that with decreasing dietary energy content a greater amount of diet is required per unit of live weight gain.

The efficiency of utilisation of metabolisable energy for gain is shown graphically in Fig. 4. The mean energy gains were 3590, 3360 and 3030 kcal. for birds on diets A, B and C respectively, and these correspond to IOM intakes of 660, 1070 and 1380 g. respectively. This would suggest that IOM in the diet had an overall effect on energy gain.

It was found from a regression analysis of energy gain on energy consumption that each kcal. difference in ME consumed, on a single diet, corresponded on average to a difference of

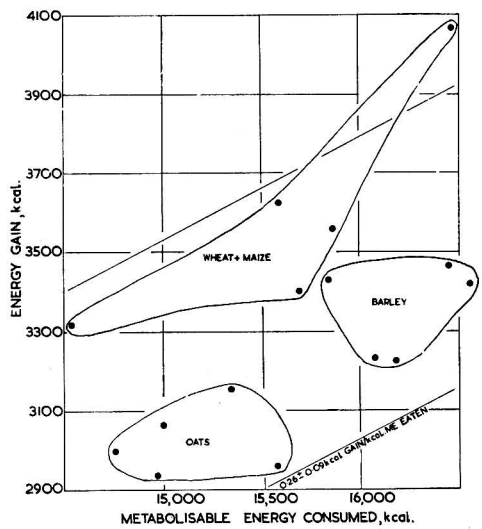

FIG. 4.—*Efficiency of utilisation of dietary metabolisable energy in Expt. 4*

0·26 (±0·09) kcal. in energy gain. This figure was used to adjust the mean energy gains on each diet to give estimates of what they would have been on an equal ME intake of 15,700 kcal. These estimates were 3610 ± 65, 3230 ± 77 and 3170 ± 79 kcal. respectively, their order being unaffected by the adjustment. The adjusted gain on diet A was significantly greater than that on diet B ($P < 0.01$) and on diet C ($P < 0.001$), but the gain on diet B did not prove significantly greater than that on diet C. This result is apparent from Fig. 4 in which the two lines above and below the data give the slope of the marginal gain. Diets giving results lying at approximately the same distance from these lines will not differ significantly in utilisation of energy, e.g., the oats and barley diets. When, however, sets of results are not equidistant from these lines, there are differences in the utilisation of dietary ME, e.g., the wheat and maize diet differs from the oats diet and from the barley diet in utilisation of energy.

One main difference in nutrient intake between the birds on the wheat plus maize diet and those on the oats or barley diets was in the intake of IOM (Table XI), which on diets B and C was about 60% and 110% greater respectively than on diet A. If such differences in IOM are the main or a contributory cause of differences in energy utilisation, then this might indicate that the energy requirements for gizzard activity and propulsion along the alimentary tract increase appreciably with large increases in the intake of IOM.

Although the utilisation of ME by birds on the oats and barley diets was practically the same, the lower final weights of birds on the oats diet are probably a reflection of the reduced intake arising from the bulk and texture of the diet.

If the quantity of IOM ingested is alone responsible for differences in utilisation of energy, then it is difficult to explain why there is no significant difference in the utilisation of energy by birds on the barley and oats diets.

It might be noted that on the oats diet C, 4% less protein was eaten than on diet A and this also might contribute to the observed difference between diets A and C.

The amino-acid contents of each diet, predicted from tables of amino-acid composition, are compared with the NRC requirements in Table XII. Only in leucine is there likely to be an appreciable difference in favour of a diet based on wheat + maize, but as the leucine contents

Table XII

Predicted amino-acid composition of diets in Expts. 4 and 5 (g. per 300 kcal. ME) as a percentage of the requirements given by the National Research Council of the United States

		Phe + Tyr	Leu	Arg	Glyc	Lys	Met + Cys	Val	Isol	Thr	Try	His
	N.R.C. requirements % for starting chicks	1·6	1·4	1·2	1·0	0·9	0·8	0·8	0·6	0·6	0·2	0·15
Expt. 4	Starting diets											
	A	112	125	116	119	142	95	145	177	131	120	347
	B	108	111	116	115	143	93	143	175	130	120	333
	C	113	114	122	111	146	95	147	181	128	135	340
	Finishing diets											
	A	102	116	99	108	124	86	131	157	120	115	313
	B	97	99	101	103	127	84	129	155	122	115	300
	C	102	102	107	98	127	85	132	163	117	120	307
Expt. 5	Starting diets											
	D	124	127	115	136	143	99	154	186	138	125	366
	E	128	132	122	128	146	101	160	193	145	135	380
	F	127	131	120	121	144	100	163	188	142	135	366
	G	111	116	100	111	124	89	139	168	125	115	333
	H	98	102	78	96	105	79	120	145	113	95	287
	Finishing diets											
	D	114	117	101	124	130	91	140	170	127	115	333
	E	117	121	107	117	129	94	143	175	130	120	340
	F	114	120	104	107	127	91	141	171	130	125	340
	G	99	106	84	99	108	81	123	150	113	105	287
	H	91	92	70	85	85	71	107	127	98	90	253

in all three diets are greater than published requirement figures it is unlikely that this amino-acid is involved in differences in utilisation of energy.

A prediction of vitamin contents of the diets from tables of averages showed that in neither fat-soluble nor water-soluble vitamins would a deficiency be expected in any of the three diets. There were differences in the content of important vitamins such as riboflavin, pantothenic acid and nicotinic acid but these were in favour of the barley and oats diets.

Comparison of the efficiency of energy conversion with (i) *increasing but low levels of dietary indigestible organic matter and* (ii) *decreasing protein levels* (Expt. 5)

In this experiment it was planned to feed diets increasing in IOM content but containing the same ratio of protein to ME, and also diets equal in IOM and ME but decreasing in protein content to levels thought to be inadequate for normal growth.

Diets

The starting diets fed to 6 weeks of age are shown in Table XIII. The amino-acid and vitamin contents of diets D, E and F were made similar to those of a diet used successfully in the Delmarva area of the United States for broiler production.[8] Diets D, E and F differed mainly in content of IOM, the ratio of crude protein to ME remaining constant. Diets E, G and H differed mainly in protein/ME ratio, the IOM remaining relatively constant. The finishing diets D–G contained 2% less white fish meal and 2% less groundnut meal but 4% more cereal than the corresponding starting diets. Finishing diet H contained 3% less white fish meal but 3% more cereal than the corresponding starting diet.

Table XIII

Starting diets for Expt. 5

Diet	D %	E %	F %	G %	H %
Wheat, coarsely ground	50	17	—	17	17
Barley, ground	20	37	43	42	47
Oats, Sussex ground	—	18	30	18	18
White fish meal	15	13	12·5	11	9
Groundnut meal	6	6	5·5	3	—
Maize gluten meal	3	3	3	3	3
Grass meal	2	2	2	2	2
Dried yeast	1	1	1	1	1
Bone flour	0·5	0·5	0·5	0·5	0·5
Ground limestone	1	1	1	1	1
Common salt[a]	0·5	0·5	0·5	0·5	0·5
Adisco[b]	0·5	0·5	0·5	0·5	0·5
Procaine penicillin mix[c]	0·45	0·45	0·45	0·45	0·45
Embazine[d]	0·05	0·05	0·05	0·05	0·05
Vitamin supplement[e]	—				
	100	100	100	100	100
ME predicted, kcal./kg.	2950	2700	2660	2760	2790
Crude protein, %	23·5	21·6	21·3	19·7	17·7
IOM (calc.), %	14	19	21	18	17
Protein/ME, g./kcal.	0·080	0·080	0·080	0·071	0·063
IOM/ME, g./kcal.	0·047	0·070	0·079	0·065	0·061

[a] Containing manganese sulphate 4 oz. in each 10 lb.
[b] Proprietary mixture containing vitamin A 1000 i.u. and vitamin D 200 i.u. per g.
[c] Containing procaine penicillin 1 g. per lb.
[d] Proprietary mixture containing 22·5% of sulphaquinoxaline
[e] To provide in the final ration riboflavin 4 p.p.m., calcium pantothenate 4 p.p.m. and nicotinamide 10 p.p.m.

Method

Day-old Rhode Island Red × Light Sussex cockerels (500) were fed coarsely ground grain for 4 days. As before, a stratum of birds chosen by weight, was randomised into 15 experimental groups of 12 birds and 1 spare group of 12 birds. These groups were randomised to diets and

pens in the same hut. The spare group was fed diet E and used as a source of replacements for birds dying during the first two experimental weeks only.

Food spillage was measured as in Expt. 4 and due allowance made in the final calculations. Bird weights and food eaten were recorded weekly.

At 12 weeks of age six birds from each group were chosen as before and killed for carcass analysis.

Results and discussion of Expt. 5

Average spillage figures were similar to those in Expt. 4, ranging from 6% to 8% during weeks 0–6, 0·3% to 1% during weeks 6–12, and 2% over the whole experimental period.

The results in Table XIV show that there were significant dietary differences in the food conversion ratios. This value went up from diet D to E ($P < 0.01$) and from E to F ($P < 0.001$), again confirming that with an increasing content of IOM, a greater amount of diet is required per unit of live weight gain. The ratio also went up from diet E to G ($P < 0.05$) and from G to H ($P < 0.01$) indicating that within these experimental limits, reduction in dietary protein content from 21½ to 19½ to 17½% in starting diets resulted in greater amounts of diet being required per unit of live weight gain.

Table XIV

Average live weights at 12 weeks and the amounts of food and certain components consumed

Diet	D	E	F	G	H	Standard error of difference
Mean final weight per bird, g.	1979	1936	1813	1875	1720	
Mean food eaten per bird, g.	5473	5631	5572	5608	5343	
Food conversion ratio, g. food/g. gain	2·84	2·98	3·16	3·07	3·19	±0·035
Protein consumed per bird, g.	1206	1148	1100	1037	869	
IOM consumed per bird, g.	770	1070	1170	1010	910	
Utilisation of ME for gain, %	20·6	21·4	21·2	21·6	18·8	±0·7

The efficiency of utilisation of metabolisable energy for gain is shown graphically in Fig. 5, which also indicates the position of the results obtained in Expt. 4 (broken line). A regression analysis showed that there was a marginal gain of 0·22 ± 0·07 kcal. for each additional kcal. of metabolisable energy consumed. This compares well with the figure of 0·26 ± 0·09 kcal. obtained in Expt. 4. The differences in efficiency of utilisation of energy among birds on diets D, E and F were not significant despite differences in intake of IOM as large as those observed in Expt. 4 (Table XI). It is perhaps worth noting however that the crude protein/Me ratios of diets D, E and F in Expt. 5 were some 10% higher than those in Expt. 4.

The utilisation of ME from diet H was significantly less than from all the other diets ($P < 0.05$) and this is reflected by the final live weights reached. This was the diet with the lowest protein/ME ratio in the starting diet, 0·063 compared with 0·080 for starting diets D, E and F and 0·071 for starting diet G. At protein/ME ratios below 0·066, which corresponds to the NRC protein requirement of 20% and an assumed dietary ME content for NRC rations of 3000 kcal./kg., amino-acid balance in the protein might be expected to become increasingly important in metabolism. Table XII shows that starting diet H is probably deficient in arginine and methionine while finishing diet H may be deficient in lysine also. Glycine also may be low in finishing diet H, but by 6 weeks of age the birds will be synthesising this amino-acid themselves. From a superficial inspection of results in Table XIV it might be argued that the birds on diet H have been unable to compensate for the inadequate dietary protein, because they have eaten less food, and have simply eaten to satisfy a requirement for energy.

In order to test this hypothesis, a study was made of the relationship between food or ME consumed and live weight. It was found that this could be described satisfactorily by the equation

$$F = aW^{b} - c \log W$$

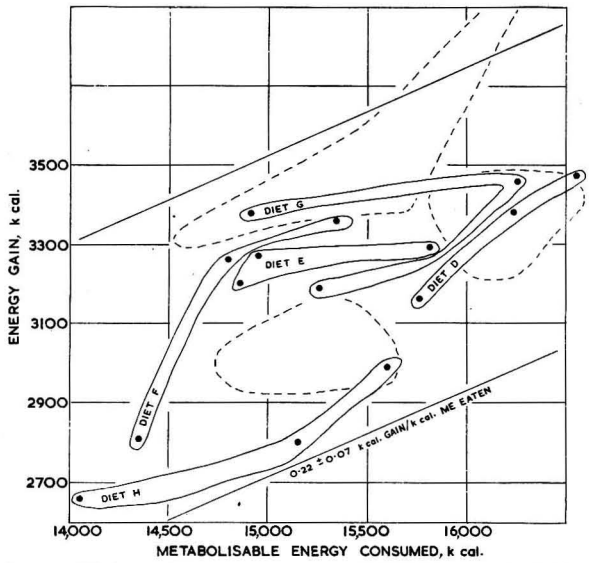

FIG. 5.—*Efficiency of utilisation of dietary metabolisable energy in Expt.* 5

where F is the average weekly food consumption per bird in g. or the average weekly ME consumption per bird in kcal., W is the estimated average weight of the bird during the week in g., a, b and c are constants and logarithms are to the base 10.

From this equation it can be seen that uptake is related to body weight raised to a power which decreases with growth of the bird.

Fifteen such equations were calculated from the results obtained in Expt. 5, one for each group of birds, but it was found they could be combined into a single equation for each set of three groups on a given diet. Further analyses showed that the same values for constants b and c could be used for each diet but values for a were different (see Table XV). The bulk of the individual values in the results from which the equations were derived fall within 12–14% of estimates made using the equations.

Table XV

Values for the constants in the equation $F = aW^b - c \log W$

Diet	Values of constant a	
	F = food intake, g.	F = ME in intake, kcal.
D	0·325	0·962
E	0·342	0·923
F	0·350	0·931
G	0·352	0·971
H	0·359	0·998

In each instance $\begin{cases} b = 1{\cdot}60\ (\pm 0{\cdot}07) \\ c = 0{\cdot}174\ (\pm 0{\cdot}013) \end{cases}$

Table XVI

Relative consumptions at any given weight in Expt. 5

Diet and characteristics				Relative consumptions		
No.	ME, kcal./kg.	Protein, % Weeks 1–6	Protein, % Weeks 6–12	Food	ME	Protein
D	2950	23·5	21·6	95	104	104
E	2700	21·6	19·9	100*	100*	100*
F	2660	21·3	19·1	102	101	100
G	2760	19·7	18·0	103	105	94
H	2790	17·7	15·7	105	109	84

* Differences not significant

On the basis of the equations and with food consumption on diet E taken as 100, the comparative consumptions of feed, ME and protein at the same live weights on diets D, F, G and H were computed and are shown in Table XVI.

Considering diets D, E and F in which the protein/ME ratio was the same and the principal differences were in the IOM/ME ratio, it can be seen that food intake on the high-energy–low-fibre diet D was relatively less than on diets E and F, but this smaller amount of food provided more ME and protein.

In diets E, G and H, in which the protein/ME ratios decreased while the IOM/ME ratios remained relatively constant, the intake of food increased per unit of live weight as the proportion of protein to energy became smaller even though this entailed the consumption of more energy than usual. However, even with these relatively larger intakes the bird's ability to compensate for inadequate protein was limited, the over-riding factor probably being an inability to lay down or get rid of the unwanted excess of energy incidentally ingested. This point is illustrated by Table XVII which shows that the birds on diet H, with a protein/ME ratio of about 0·06, disposed of considerably more heat per unit of live weight than birds on the other rations. This is also true when the calculation is made on a surface area basis ($W^{0·7}$).

Table XVII

Disposal of heat energy (Expt. 5)

Ration no.	Protein/ME ratio in starting ration	kcal. ME lost as heat during experiment	Average live wt. (W) of bird during experiment, g.	kcal. ME disposed as heat per g. average live wt. during experiment	$W^{0·7}$	kcal. disposed as heat per unit of $W^{0·7}$ during experiment
D	0·080	12,830	852	15·0	112·6	114
E	0·080	11,980	811	14·8	108·7	110
F	0·080	11,660	767	15·2	104·6	111
G	0·071	12,160	789	15·4	106·7	114
H	0·063	12,150	694	17·5	97·5	125

It may be then, that birds can and do compensate within limits for low or poor quality protein in the ration, i.e., they can compensate to some extent for a low protein/ME ratio. In order to do so, however, they consume more than a normal amount of energy and have to dispose of this extra energy as heat. Their limited ability to make this disposal may be one limiting factor in the uptake of feed which contains inadequate protein.

That birds eat for protein as well as energy when the former is below the optimum for the diet, is in agreement with observations by Donaldson *et al.*[9] who stated moreover that ' much of the additional energy consumed was deposited as fat in place of water in the carcass '. This general statement may apply to their types of diet in which the dietary protein/ME ratio was lowered by adding fat, but does not apply in our Expt. 5, because no appreciable difference could be found between groups in the carcass composition of the birds (Table XVIII). The apparent disagreement with the American results may arise from an inherited difference in the ability of birds to dispose of excess dietary energy.

Table XVIII

Carcass analysis of birds at end of Expt. 5

Diet	D	E	F	G	H
Final weight per bird (mean), g.	1979	1936	1813	1875	1720
Water, g.	1380	1349	1246	1286	1207
Protein, g.	412	405	394	399	352
Fat, g.	127	117	112	133	107
% Protein in carcass	20·8	20·9	21·7	21·3	20·5
% Fat in carcass	6·4	6·0	6·2	7·1	6·2

Work on this aspect of the problem is now being carried out.

Conclusions from Expts. 4 and 5

The contents of IOM in the rations tested ranged from 12 to 24%, covering the range normally expected in practice. There was no consistent effect of IOM on energy utilisation

although at the lowest level a wheat plus maize ration proved superior to all others tested. This effect does not seem to be due entirely to its low content of IOM.

At the highest level, a ration containing oats only as the cereal, gave poor growth although conversion of ME was normal. The poorer growth was attributed to reduced intake owing to the relative unpalatability and bulk of this ration.

Ration H containing a medium level of IOM of about 17% and a critical ratio of protein/ME in the starting ration of about 0·06 also gave rise to poor growth. This ration was probably deficient in arginine and methionine (Table XII) and the birds tended to compensate for the amino-acid deficiency by consuming more food and thus energy than was normal for their weight and to eliminate the excess energy as heat.

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Rowett Research Institute
Bucksburn
Aberdeen

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THE EXPRESSIBLE FLUID OF FISH FILLETS. X.*—Sodium and Potassium Content in Frozen and Iced Fish

By R. M. LOVE

Sodium and potassium were determined in the expressible fluid of thawed fish which had been frozen at different rates. The sodium level altered little, but the potassium rose and fell at certain rates, corresponding to the variations in the concentration of protein. No new information was obtained on the mechanism of the freezing process, but the results were considered to support the thesis that at least part of the intracellular potassium is not free to diffuse, even after death. The ash content of the expressible fluid from fish kept for various times in ice showed little change in the proportion of sodium, but potassium decreased over the first 4 days and increased again after about 14 days.

Introduction

Previous papers in this series have reported analyses of the fluid obtained by applying pressure to cod fillets which previously had been frozen and then thawed. Since there was evidence that the expressible fluid was mainly extracellular in origin,[1] any increase in the concentration of intracellular constituents was taken to mean that the cells had been split open during freezing. Analyses of deoxyribose nucleic acid (DNA), solids, protein nitrogen, ash

* Part IX: *J. Sci. Fd Agric.*, 1958, **9**, 262.

and mitochondria were carried out and, chiefly on the evidence of the DNA determinations, it was deduced that ice would damage the muscle cells during freezing under three quite narrowly-defined sets of conditions. These were when the fillet centres froze from 0° to −5° in about 25 min. (' zone A '), 80 min. (' zone B ') and 200–500 min. (' zone C '). With the aid of photomicrographs, the mechanisms whereby the ice damaged the cells in these three zones were worked out.[2–5]

This present note summarises the results of analyses of two mineral constituents. It is well known that most of the sodium in living muscle is situated in the extracellular space, while the potassium is intracellular. It was hoped therefore that by studying the relative concentrations of these substances some further light might be thrown on the nature of the freezing process.

Experimental

Material

Cod (*Gadus callarias* L.) between 18 and 22 in. long were used, which had been caught in the North Sea about 30 miles from Aberdeen. They were gutted at sea and brought to the laboratory packed in ice.

Methods

The technique of freezing fillets at a number of different rates has been described previously.[3] The expressible fluid, obtained as before[2] was evaporated to dryness at 100°, then dry-ashed in an electric furnace at 550°. Sodium and potassium were determined with a Hilger Uvispek spectrophotometer with a flame attachment, at wavelengths of 589 and 766·5 mμ respectively. Total solids were found by weighing the residue after a sample of expressible fluid had been dried at 100° to constant weight.

Results

Expressible fluid of fish frozen at different speeds

Three experiments were carried out in which cod fillets, from fish kept respectively 4, 4 and 6 days in ice, were frozen at eight different freezing times. After the fish had been allowed to thaw, the expressible fluid was analysed for total solids and for sodium and potassium.

It was found that changes in the level of solids were followed closely by those of potassium, as in Fig. 1 which illustrates one of the experiments. There did not appear to be any correlation between solids and sodium.

Unfrozen fish stowed in ice

Cod were stowed in ice at an ambient temperature of about 2°, four fish being removed at intervals for filleting and submitting to a pressure of 7 lb./sq. in. for 3 h. It was found that the total solids in the expressible fluid fell from almost 9% (2 days in ice) to 5·4% (21 days), and that there was a similar fall in ash content, illustrated in Fig. 2.

The Figure shows that the ash content of the expressible fluid of newly killed fish is higher than that of whole cod muscle (about 1·2%) and that it falls steadily during subsequent stowage of the fish in ice. When the two constituents of the ash were analysed on a basis of weight per unit volume of expressible fluid, they were found to decrease steadily with time in ice. However, on a ' % of total ash ' basis it was seen that sodium values remained fairly constant at about 9%.

Potassium values behaved differently. Between the 1st and 4th days there was a fall, and then a gradual rise after about the 14th day. In order to check this finding, three further similar experiments were carried out, only potassium being determined. In all four experiments the initial drop in the proportion of potassium was observed, and the subsequent rise was shown in three of them. The exception showed a rise on the 16th day, but the final point at 22 days was down again—perhaps a chance variation.

Two of the experiments are illustrated in Fig. 3. The single high value at 11 days in the upper experiment was not seen elsewhere and so is probably not significant.

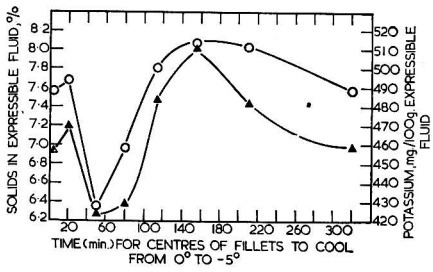

FIG. 1.—*Comparison of the solids content* (○), *i.e., the residue after heating at* 100°, *with the potassium content* (▲) *of the expressible fluid of cod fillets which had been frozen at different speeds and then thawed*

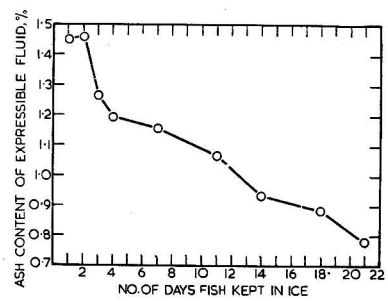

FIG. 2.—*Ash content of the expressible fluid of cod fillets after the whole gutted fish had been stowed in ice for various times*

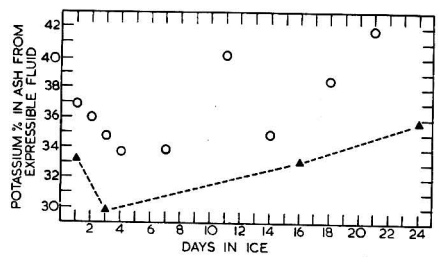

FIG. 3.—*Potassium content of the ash from the expressible fluid of cod fillets obtained from whole gutted fish stowed in ice for various times*
(two experiments)

Discussion

As regards information on the mechanism of freezing, clearly we do not learn anything new by analysing expressible fluid for sodium and potassium. From Fig. 1, it appears that potassium analyses run parallel to 'solids', and so could give some information on cell damage in fillets frozen at different speeds. 'Solids' are however less reliable[2] as an indication of cell damage than DNA concentrations, so that there is no advantage to be gained from carrying out potassium analyses.

It is noteworthy that the potassium does not diffuse freely from the cells, even several days after the death of the fish, but only escapes after the cells have been ruptured.

Simon *et al.*[6] considered that part of the potassium in toad muscle was bound and part free to diffuse, all being intracellular. This thesis appears to find support in the results illustrated in Fig. 3, where the fish were stowed in ice. The small drop in the first 4 days of stowage in ice probably reflects the diffusion and leaching out of most of the 'free' potassium, while the subsequent rise at about the 14th day shows cell rupture and protein liberation by bacterial action—protein liberation bringing 'bound' potassium into the extracellular spaces. There is a parallel here with some previous work,[1] in which the DNA concentration of expressible fluid of cod fillets was measured after the intact fish had been stowed for various times in ice. It was found that if the skins of the fish were thoroughly washed in dilute formaldehyde to kill the bacteria, the expressible fluid DNA showed no change over a 23-day period, but that the DNA level in unwashed fish showed a sudden steep rise at about 14 days, which was considered to be the result of bacterial action breaking the cells open.

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Dept. of Scientific & Industrial Research
Torry Research Station
Aberdeen

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THE EXPRESSIBLE FLUID OF FISH FILLETS. XI.*—Ice Crystal Formation and Cell Damage in Cod Muscle Frozen before Rigor Mortis

By R. M. LOVE and S. B. HARALDSSON†

Fillets of cod were frozen at different speeds, both before and after the onset of rigor mortis, and the ice crystal patterns and cell damage were studied. In pre-rigor fish the intracellular ice crystals were smaller than those in comparable post-rigor fish, and also intracellular ice was seen in pre-rigor fish at much slower freezing rates than in post-rigor material. Cell damage ' peak B ' (as measured by the deoxyribose nucleic acid method) which usually occurs in post-rigor fillets at a freezing time of about 80 min. was found to be at about 200 min. in pre-rigor fillets. Possible mechanisms are discussed.

Introduction

A study has been made in this series of the way in which ice grows during the freezing of cod (*Gadus callarias* L.) muscle, and how the muscle cells can be damaged during the process.[1–4] Histological techniques were employed, and also a new technique devised for the purpose[1] in which, after the fillets had been thawed, the concentration of deoxyribose nucleic acid phosphorus (DNAP) in the expressible fluid was analysed and taken as a measure of the number of cells burst open. The technique revealed that there were three rates of freezing which resulted in maximum cell damage, namely when the temperatures at the centres of the thickest parts of the fillets dropped from 0° to −5° in about 25 min. (' zone A '), about 80 min. (' zone B ') and 200–500 min. (' zone C ').

All fish used in those experiments had been kept in ice for at least one day after being caught and so had passed through rigor mortis. The present paper describes a study of fish frozen before this stage (' pre-rigor fish '). The work was done in order to find out whether the same phenomena occurred in the pre-rigor fish or if, for instance, the extra resilience characteristic of pre-rigor cells[5] enabled them to withstand the action of the ice.

Material and methods

The raw material and the technique for freezing fillets at different speeds have been described previously.[2]

* Part X: preceding paper
† Present address: c/o The Fisheries Association of Iceland, Skulagata 4, Reykjavik, Iceland

Results

Experiment 1

Conditions of experiment.—The main difficulty in this type of investigation is that cod generally go into rigor mortis (become rigid) within 3–6 h. of being caught, so that the time left to work in is very short. The first experiment was therefore carried out at sea on the research trawler *Sir William Hardy*, NE. of the Faroe Islands on the 7th August, 1957. An aluminium tank was made, 3 ft. × 1½ ft. internal dimensions, 1 ft. deep, having insulated sides. The bottom was double, enclosing a chamber 5 in. deep, which was filled with ethanol cooled to −78° with solid carbon dioxide, the latter being fed down a wide-necked funnel welded on to the side of the enclosed chamber under the tank. The movement of the ship swirled the alcohol about and ensured that the entire floor of the box was kept below −60°. The apparatus was a non-spill modification of the one used in the laboratory.[2]

Controls (post-rigor) were brought from Aberdeen. These consisted of cod caught on a commercial boat, stowed for 5 days in ice and filleted just before the departure of the *Sir William Hardy*. They were kept a further 2 days at chill temperatures in the fish room of the research ship, and then frozen along with the pre-rigor fillets.

At the first haul, fillets from 18 newly-killed cod and 6 control fillets were placed with the cut surface downwards on the freezing surface, i.e., the bottom of the tank. A thermocouple was tied in place in the centre of the thickest part of each of two control fillets, and cooling curves were plotted by a Honeywell–Brown automatic recording potentiometer.

An aluminium plate 2 mm. thick was placed on top of all the fillets, and a second layer of 36 pre-rigor, 6 post-rigor fillets made up in the same way. Two further similar layers were built up, and then the top of the tank was covered with insulating material. Only four layers were made from the fish of the first haul because each successive layer freezes more slowly than the one below it, and it was feared that rigor mortis would occur in any further layers before they had time to freeze. In fact, as will be seen, the fourth layer did go into rigor before freezing commenced.

A fifth layer was added 10 h. later from a new haul, and a sixth layer after another 26 h. The fillets were then separated, wrapped in aluminium foil, and stored at −45° until the end of the trip (15th August, 1957).

Testing for pre-rigor freezing.—Frozen fillets which contracted appreciably on rapid thawing were regarded as pre-rigor frozen; if there was no contraction, it was considered that freezing had occurred after rigor had set in.

Four fillets from each layer were measured and subsequently thawed by placing in thin polythene bags in water at 30° (thawing time about 10 min.). They were then measured again, and the average difference in length, expressed as a percentage of the 'frozen' length, is shown in Table I.

Table I

Characteristics of cod fillets frozen pre-rigor at different rates at sea, August 1957

Layer in freezing box	Time (min.) taken for centres of fillets to cool to 0° from moment of landing on deck	Time (min.) taken for centres of fillets to cool from 0° to −5° ('freezing time')	Average shortening when thawed in warm water, as % of frozen length
I	33	30	16·0
II	148	96	21·4
III	309	135	4·5
IV	392	410	1·0*
V	198	445	—*
VI	90	180	10·0

* Layers IV and V were not used in the subsequent investigations, being considered from these figures to have gone into rigor before freezing.

DNAP determinations.—The six control fillets in layers I, II, III and VI were thawed out in air at 18° and submitted to a pressure of 7 lb./sq. in. at 2° in the manner already described,[1]

and 3 h. later the expressed fluid was collected and analysed.[1] The concentration of DNAP against freezing time is shown in the top curve of Fig. 1, where a typical ' peak B ' can be seen at a freezing time of 96 min. This peak marks the changeover from intracellular to extracellular ice formation.[2]

Six pre-rigor fillets were taken from each of the same layers and thawed in the same way (about 5 h.). A further six were thawed in polythene bags in water at 30° (about 10 min.). The DNAP curves corresponding to these two sets of conditions are shown at the bottom of Fig. 1.

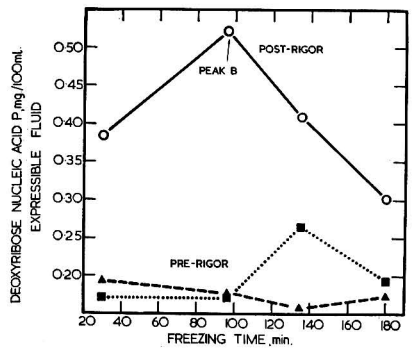

FIG. 1.—*Concentration of deoxyribose nucleic acid phosphorus in the expressible fluid of thawed cod fillets frozen at different speeds (Experiment 1), frozen at sea 1957*

○——○ Post-rigor controls, thawed 5 h. in air at 18°
△----△ Pre-rigor, thawed 5 h. in air at 18°
□ · · · · □ Pre-rigor, thawed 10 min. in water at 30°

Two facts are at once apparent in both sets of pre-rigor fish: firstly the amount of cell damage as measured by this technique was much less than in the control at all rates of freezing, and secondly peak B had disappeared. A small peak at 135 min. was present in one of the groups of pre-rigor fish, but as will be seen in the histological appraisal it was probably not significant.

Histological examination.—Sections of the frozen material were prepared after subliming off the ice as described previously,[1] and it was clear that the ice crystal patterns of pre- and post-rigor fish frozen under identical conditions were not the same.

In the samples frozen in 30 min., both pre- and post-rigor fish showed intracellular freezing, but the ice crystals in the pre-rigor samples were smaller (not illustrated). At 96 min. the controls showed extracellular freezing with some signs of cell damage (Fig. 2A) and appeared to be somewhat beyond the peak B maximum. The ice in the pre-rigor fish (Fig. 2B) on the other hand was all intracellular, and there was little sign of cell damage. These pictures correlated well with the DNAP results and, in the controls, with previous experience.[1, 2, 6]

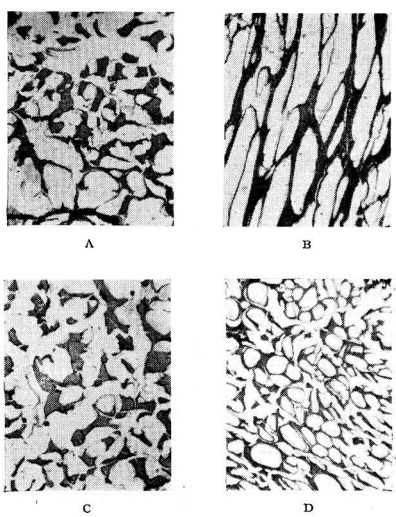

FIG. 2.—*Sections of frozen fish relating to Experiment 1*
(Ice shows white, cell material dark)
Post-rigor: frozen in (A) 96 min. (C) 180 min.
Pre-rigor: „ „ (B) 96 min. (D) 180 min.
1 mm.

The photographs in the lower part of Fig. 2 illustrate the most slowly-frozen fish in the experiment, freezing time 180 min., well beyond peak B in the controls (upper part of Fig. 1). The photograph of post-rigor fish is exactly as normally expected at 180 min.—all the ice extracellular, not much aggregation of the now dehydrated cells, little cell damage. In complete contrast, the pre-rigor sample shows all-intracellular ice—the appearance is that normally associated with post-rigor cod frozen in 50–65 min.; peak B has not even been reached.

The importance of these results made it necessary to repeat the work, if possible to extend the lengths of the freezing times and find out if pre-rigor fillets ever showed the ' peak B ' type of damage. A further experiment was therefore performed by the senior author in December, 1959.

Experiment 2

Conditions of experiment.—This work was carried out in the laboratory with live cod from an aquarium, freezing being as described earlier[2] except that an all-aluminium box was used which was larger than that in previous experiments, enabling 10 fillets to be frozen at each speed (only 4 could be frozen with the earlier apparatus).

The live fish had been caught by trawling about 20 miles E. of Aberdeen 4–6 days previously and rested in large aquaria in the intervening time. Controls were caught 30 miles SE. of Aberdeen on another ship, being gutted at sea and stowed in ice for 5 days before the experiment. All fish were about the same size, intact body lengths ranging from 19 to 22 in.

Five fillets from newly-killed fish, two containing copper–constantan thermocouples in the middle of the thickest part, and five post-rigor fillets, two with thermocouples, were placed side by side, skin uppermost, on the bottom of the freezing box. Two sheets of aluminium foil 0·025 mm. thick were laid on top, and a further similar layer of pre- and post-rigor fillets was added. A sheet of aluminium 3 mm. thick was laid on top of the second layer, and weights totalling 7 kg. distributed about the surface, in order to squash the fillets slightly and ensure good contact with the bottom of the box. ' Cellosene ' wadding was then placed on top as an insulator.

The box was then immersed to the extent of 1–2 in. in a tank of ethanol cooled to −78° with solid carbon dioxide. The temperatures from the four thermocouples in each layer (2 pre-rigor, 2 post-rigor) were plotted by a Honeywell–Brown automatic recording potentiometer.

When the temperature of the middle of the second layer reached 0° (after 45 min.), the insulation, weights and metal sheet were removed and two more layers of fish were added, the fish for pre-rigor fillets being newly killed for the purpose. The weights and insulation were then replaced. Two hours later a fifth layer was added, and the weights reduced to 5 kg. A sixth layer was added after a further 3 h., and covered only with insulation but no weights.

By the following day all layers were below −60°. The box was transferred to a cold-room at −29° and the frozen layers separated. Small pieces were cut off each fillet with a hand-saw as described[1] and the ice was sublimed under vacuum at −29°.

DNAP determination.—The rest of the material was thawed out and submitted to pressure as in the 1st experiment above. The concentration of DNAP in the expressible fluid is shown in Fig. 3, plotted against the freezing time; it can be seen that a bigger range of freezing times was obtained than in the 1st experiment.

Each pre-rigor group had a shorter freezing time than its corresponding post-rigor group—for instance the pre-rigor fillets in the top layer cooled from 0° to −5° in 290 min., as against 380 min. for the post-rigor fillets. Now the post-rigor fillets, having been stored for a few hours in a chill-room, were all at about 2° when placed in the freezing box, while the pre-rigor material, straight from the aquarium, was at a higher temperature (about 12°); Long[7] has shown that the higher the initial temperature is above the freezing point, the quicker the fish pass from 0° to −5°, other things being equal, so the present observation is probably an example of this phenomenon, which has been explained thermodynamically.[7] Amano *et al.*[8] however found that pre-rigor fillets cooled more slowly below the freezing point than comparable post-rigor fillets, which they explained on the grounds that some heat would be liberated during the freezing of pre-rigor muscle by the breakdown of adenosine triphosphate, absent from the

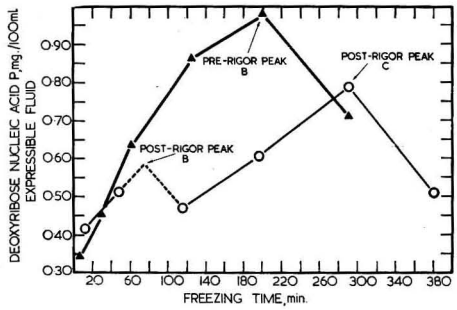

FIG. 3.—*Concentration of deoxyribose nucleic acid phosphorus in the expressible fluid of thawed cod fillets frozen at different speeds in the laboratory (Experiment 2, 1959)*

The identity of 'peak B' is established from the histological sections in Fig. 4

△——△ Pre-rigor
○——○ Post-rigor

post-rigor fillets. If such a phenomenon occurred at all in the present work it was completely masked by the presumed 'initial temperature effect' noted above.

In the post-rigor curve, Fig. 3, there are well-marked peaks B and C in the usual locations—see introductory section. As stated previously[2] it is fortuitous if an experimentally determined point coincides exactly with a maximum DNAP value, because the freezing times of the different layers depend on the thickness of the fillets; it can be seen that there is here no point exactly at peak B. The histological sections showed that the ice at 11 min. (post-rigor curve) was all intracellular (Fig. 4A), at 47 min. mostly intracellular with some cell damage (4B) and at 115 min. all extracellular (4C). The true peak B therefore does lie somewhere between the latter two points.[2]

The pre-rigor curve has just one high peak at 200 min. The remarkable fact shown by the histological sections is that it is in fact peak B and not C as might have been supposed. Unlike in Fig. 1, the pre-rigor DNAP level at about 80 min. is higher than that of the post-rigor DNAP, but this is doubtless the influence of the very high pre-rigor peak B in the second experiment—the heights of the peaks vary greatly in different experiments.[2]

Histological examination.—When the sections of the quickest-frozen fish are compared, three facts stand out: in the pre-rigor fish (Fig. 4D) the ice is lobulated (rosette-shaped) not roughly circular as in post-rigor fish (4A), the ice crystals are smaller, and there is actually a smaller proportion of ice to tissue.

Taking the ice patterns near the peak B maximum, it can be seen that in pre-rigor fish (4E) virtually all the freezing is intracellular at a freezing time of 200 min., while the proportion of extracellular freezing in the post-rigor fish (4B) is already considerable at 47 min. in this experiment. Beyond peak B, all freezing is extracellular in the post-rigor fish (4C) frozen in 115 min., while at 290 min. pre-rigor (4F) there is still some intracellular freezing, although the greater part of the ice has formed extracellularly.

While the disposition of the ice in the slowest frozen pre-rigor group was different from that in the other pre-rigor groups, the *amount* of ice measured histologically was the same, being considerably less than in the post-rigor groups (see Discussion). It is improbable therefore that rigor had set in in the pre-rigor fillets frozen in 290 min.

Two blocks of fish were sectioned from each of the five pre-rigor and post-rigor fillets frozen at each speed: the other sections which have not been illustrated support the same conclusions. Some of the slowest-frozen pre-rigor sections showed all-extracellular freezing, some showed mostly intracellular freezing: Fig. 4F is noteworthy in illustrating the two types together, sharply demarcated.

Thus, as Piskarev *et al.*[10] have concluded, the concept of quick and slow freezing as defined by the size or location of ice crystals is meaningless unless the physiological state of the muscle is taken into account.

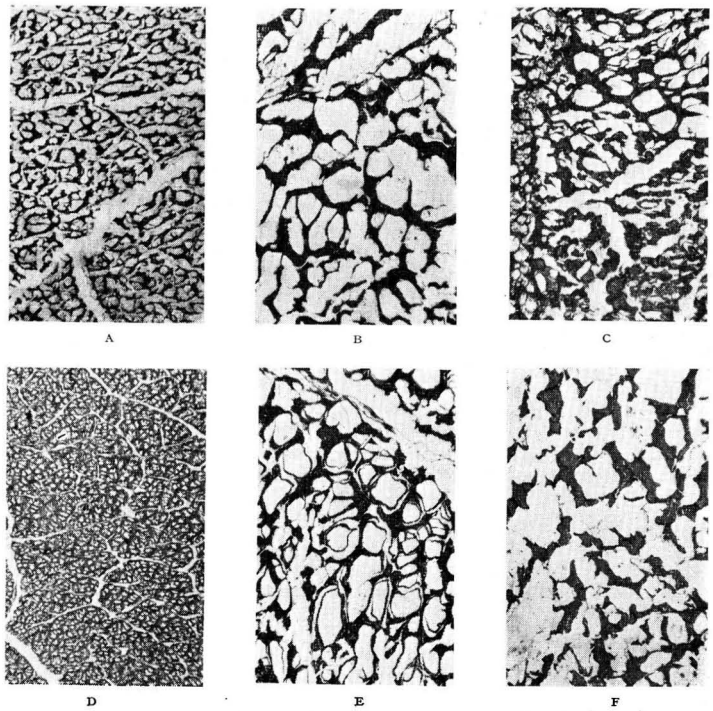

FIG. 4.—*Sections of frozen fish relating to Experiment 2*

Pictures B and E correspond with ' peak B ' in post- and pre-rigor fish, respectively, but note that the freezing times are very different

Post-rigor: freezing time, min. A 11, B 47, C 115
Pre-rigor: " " " D 6½, E 200, F 290

1 mm.

Discussion

The experiments show that the pattern of ice-crystal formation in pre-rigor and post-rigor fish is not the same. Concurrently with a preliminary report of the work[9] there appeared a paper by Piskarev & co-workers[10] describing similar effects in fresh-water fish. They found that freezing pre-rigor muscle at −25° resulted in small intracellular ice crystals, while if the same material were held for 6 days in ice before freezing (post-rigor), the ice crystals were large and extracellular.

A complete explanation for the phenomenon necessitates a better knowledge of the mechanism of the growth of ice-crystals in biological systems than at present exists, but it is possible at this stage to suggest some alternatives.

Intracellular freezing

Lusena & Cook[11] have pointed out that, in the slow freezing of biological tissue, the ice phase is essentially continuous as water moves through the tissues to support crystal growth. In rapid freezing, however, the ice phase is discontinuous, perhaps because the rate of crystal growth in the tissue exceeds the rate of moisture movement.

In the present work it has been shown that where there is intracellular freezing, the ice crystals in pre-rigor muscle are more numerous than those in comparable post-rigor muscle,

i.e., they are ' more discontinuous '. The explanation may be that in pre-rigor muscle a greater proportion of the water is combined with the protoplasmic jelly, thus slowing down the rate of the movement of free water, and so of ice-crystal growth. Since the zone of low temperature still proceeds apace into the tissue, new ice nuclei are caused to form ahead of the growing ice front so that the ice pattern finally appears as a number of discrete accretions around nuclei, not of continuous shafts or spears. The onset of rigor mortis is marked by a shrinkage of the muscle cells[12] and the exudation by them of a watery fluid into the extracellular space. It seems reasonable to think that when the myofibrils within the cells shrink, some of the newly-liberated water remains within the cells amongst the myofibrils and greatly facilitates crystal growth, so that the ice can now form larger more continuous bodies. It is reasonable also to relate the lobular shape of the ice (increase of surface area) in pre-rigor tissue—Fig. 4D—to the lowered accessibility of the freezable water.

Supporting evidence for this concept is to be found in a quantitative estimate of the amount of bound water, which is to be given as a separate study,[5] and which reveals that the water bound by the protein at low temperatures is considerably greater in pre-rigor cod muscle than in post-rigor at all rates of freezing. The phenomenon can be observed in Fig. 4D (pre-rigor) where there is patently a greater area of tissue relative to ice than in 4A (post-rigor). It is easy to visualise a slowing-down of water diffusion and so of crystal growth under such conditions.

The alternative explanation rests on differences in heat production in pre- and post-rigor ice crystallisation. Most if not all biological tissues supercool. When the temperature becomes sufficiently low, nucleation occurs, ice forms rapidly and the temperature rises with the heat of crystallisation. If sufficient heat is generated, ice growth stops, the crystal being preceded by an accumulation of relatively warm material. In the observations of Rey-Dhaussy & Rey,[13] who used supercooled water in narrow glass tubes seeded from one end, thin finger-like ice processes grew round the edge of the warm material and seeded the supercooled water beyond, so enabling crystallisation to continue further down the tube after an interruption. Now, as already noted, Amano *et al.*[8] attribute the slower freezing of pre-rigor muscle to some additional heat produced from the breakdown of ATP, etc. If this heat were liberated immediately in front of the advancing ice, it is conceivable that a greater discontinuity in the ice crystals would result. This explanation is less likely than the first, however, since it requires most of the thickness of the fillet to be supercooled before freezing commences.

Extracellular freezing

As far as present knowledge goes, freezing in post-rigor fish can be considered to start in the watery extracellular fluid. Since the salts present in dilute solution are not incorporated into the growing ice, they gradually become concentrated and are driven on ahead of the ice front.[11] If freezing is fairly rapid, the temperature of this solution drops several degrees, the freezing point being much depressed by the solutes in it, and being in contact with cell walls on all sides it is able to cool the cells down to the point where intracellular ice can start to form spontaneously, without direct contact with the extracellular ice.

In slow freezing, the temperature of the concentrated solutes does not drop as much, and ice therefore cannot form within the cells. Instead, water diffuses out through the cell walls to the concentrated solutes, diluting them and later augmenting the extracellular ice. The cells become dehydrated until they can no longer freeze internally even at very low temperatures.

Experiments 1 and 2 have shown that intracellular ice forms much more easily in pre-rigor than in post-rigor fish, so that it is necessary to freeze very slowly in order to obtain extracellular freezing. One explanation for this is the same as for intracellular freezing, namely that since there is more bound water in pre-rigor muscle, the diffusion of free water outwards through the cell wall is slowed down, and that freezing must be very slow in order to give the free water time to leave the cell.

There is however another possibility. This is that the very small amount of extracellular fluid found in pre-rigor fish (non-existent according to Piskarev *et al.*[10]) is more concentrated with respect to dissolved material than is the more copious post-rigor fluid. If the solutes depress the freezing point of this extracellular fluid below or equal to that of the cell contents, then the first freezing could occur intracellularly, not extracellularly as in post-rigor material.

Evidence that the extracellular fluid becomes less concentrated some time after death is shown in the fact that the freezing point of tissue generally rises after death.[14] Otake,[15] who studied fish muscle, found that the freezing point rose steadily from about −3° to about −1° during the post-mortem period, and observed that 'ice production was difficult at the early stage'.

Unfortunately it has never been possible to isolate pre-rigor extracellular fluid for freezing point measurements although fluid can be obtained from post-rigor cod fillets which appears to be mainly extracellular.[16]

Thus a number of facts still remain to be found out before the full explanation for these phenomena can be reached.

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Dept. of Scientific & Industrial Research
Torry Research Station
Aberdeen

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KAFFIRCORN MALTING AND BREWING STUDIES. VIII.*— Nutritive Value of some Kaffircorn Products

By M. C. AUCAMP, J. T. GRIEFF, L. NOVELLIE, B. PAPENDICK, H. M. SCHWARTZ and A. G. STEER

Protein, fat, fibre, ash, thiamine, riboflavin and nicotinic acid were determined in eight samples each of kaffircorn (*Sorghum vulgare*) grain and malt and in one sample of breakfast food prepared from these.

Twenty-one samples of kaffir beer produced on a large scale in municipal breweries were analysed, particular attention being paid to their content of B vitamins. Changes in the vitamin content during fermentation were studied. Other factors influencing the vitamin content of the beer are discussed.

Introduction

The production of kaffircorn or grain sorghum (*Sorghum vulgare*) in the Union of South Africa varies between 100,000 and 300,000 short tons annually. In normal years the bulk of the crop is used locally, the surplus being exported to Europe where it finds its way mainly into animal feeds. The use of kaffircorn in South Africa is unique in that nearly 90% of the total goes into the brewing of kaffir beer, the traditional drink of the Bantu[1] (Table I). Most

* Part VII: *J. Sci. Fd Agric.*, 1960, **11**, 467

Table I

Production and use of kaffircorn in South Africa

(short tons)

	1955/56	1956/57	1957/58	1958/59
Production	154,800	181,100	219,500*	265,900*
Use				
Domestic				
Human consumption				
Not commercially processed	4800	6600	9100	14,800
Processed to meal, flour, etc.	7000	7500	9700	9300
Beer production	129,600	151,300	158,700	179,900
Animal feeds	2000	2500	2400	2600
Seed	2900	2900	3100	3100
Total	146,300	170,800	183,000	209,700
Export	800	1300	38,200	56,500
Waste and loss	7700	9000	9600	11,000

* Estimates

of this is used for home brewing since purchases by municipal breweries represent only 12–15% of the whole, in spite of the steep rise in municipal beer production in urban areas in recent years. This is partly due to the fact that there has been an increasing tendency to use maize products instead of unmalted kaffircorn grain in brewing on a large scale, many breweries today using kaffircorn only in the form of malt (Table II).

Table II

Use of kaffircorn by municipal breweries

Year	Beer production (millions of gal.)	Raw materials used (short tons)		
		Kaffircorn grain	Kaffircorn malt	Maize products
1954/55	25·6	7710	15,569	5828
1955/56	30·2	4538	17,204	10,704
1956/57	34·8	5892	16,341	12,212
1957/58	40·5	5139	18,100	14,005
1958/59	44·0	3566	19,669	19,860

Besides that taken in the form of beer, a further 8–9% of the domestic use of kaffircorn is for human consumption, either without any prior treatment or after processing into breakfast or baby foods. Animal feeds account for less than 1% of the crop, whilst none is processed to starch or other industrial products. Grain sorghums are also used extensively for brewing in other parts of Africa, as well as in China[2] but, in contrast to the Union, the greater part of the crop in these countries is consumed in the form of unfermented foods. In the United States of America grain sorghums are grown chiefly for animal feeding, although a certain amount is processed to starch for food and industrial uses.

As part of our studies on the use of kaffircorn in the brewing of kaffir beer in South Africa, the nutritive value of the grain and of the malt and beer produced from it have been determined. A considerable amount of work on this subject was done in the forties by Golberg and his co-workers[3, 4] but the beers which they examined were either home brewed or were municipal or mine beers brewed on a small scale under rather primitive conditions in the early days of the industry. The present work was undertaken primarily to provide figures for beer as brewed on a large scale at the present time.

Experimental

Materials

Typical samples of kaffircorn of the short red, white, birdproof, Martin and Hegari varieties were obtained from the Potchefstroom Agricultural College. The remaining samples were mixed grain of commercial origin.

The malts were commercial samples prepared in some cases by the open floor process and in others by the indoor pneumatic method.[1]

The beers were collected from municipal breweries on the Witwatersrand and in Pretoria. In order to stop further fermentation, they were chilled in dry ice during transport to the laboratory and were stored at 0° or below until analysed.

Methods

Moisture, nitrogen, fat, ash and fibre in the grains and malt were determined by the methods of the A.O.A.C.[5] as was alcohol in beer. Total solids in beer were determined by evaporation to dryness on a water-bath followed by heating in an oven at 103° for ½ h. Nitrogen in the beer was determined by a modification of the method of Ma & Zuazaga.[6] Soluble solids and nitrogen were determined on the clear supernatant obtained after centrifugation of the beer.

The B vitamins were determined microbiologically, thiamine according to Snell,[7] nicotinic acid according to Snell[7] and Barton-Wright[8] and riboflavin according to the British Pharmaceutical Codex.[9]

Ascorbic acid was determined by a modification of the indophenol method of the British Pharmacopoeia.[10]

Results and discussion

Composition of grain and malt

Analyses of eight samples each of grain and malt are summarised in Table III. Figures for the amino-acid composition of three of the grain samples and one malt are reported in the following paper.[11] Unpublished work in this laboratory has shown that the nitrogen content of kaffircorn can vary over a much wider range than is reported here, viz., from 1·22 to 2·47% (7·6–15·4% crude protein). More detailed studies of the nitrogenous constituents of the grain and their changes during malting and brewing are in progress.

Table III

Composition of kaffircorn, kaffircorn malt and a breakfast food prepared therefrom

(all results on dry weight basis)

	Grain (8 samples)		Brewers' malt (8 samples)		Breakfast food (1 sample)
	Range	Mean	Range	Mean	
Protein (% N × 6·25)	8·5–11·9	10·7	10·0–12·2	11·0	9·5
Fat (%)	1·6–4·6	3·6	1·4–3·6	2·8	3·6
Fibre (%)	3·4–7·3	4·7	2·5–5·0	4·2	3·5
Ash (%)	1·4–2·2	1·7	1·3–2·0	1·6	3·0
CaO (as % of ash)	2·1–4·1	3·1	0·9–4·6	2·6	2·9
P_2O_5 (as % of ash)	31·6–47·7	40·7	36·1–51·0	44·6	24·1
Thiamine (μg./g.)	4·6–6·8	5·8	3·2–8·5	5·1	7·0
Riboflavin (μg./g.)	1·2–3·1	2·0	2·0–3·4	2·6	1·7
Nicotinic acid (μg./g.)	23 – 40	32	35–47	41	35

The values for nicotinic acid and riboflavin found in the present study are very similar to those previously reported for kaffircorn grain and malt by Golberg & Thorp[4b] but our values for thiamine tend to be higher than theirs which averaged 3·0 μg./g. (range 2·1–4·9) for grain and 1·7 μg./g. for malt.[4a, 4b] In general, kaffircorn and kaffircorn malt are better sources of riboflavin and particularly of nicotinic acid that is whole maize which contains 0·7–1·6 μg. of riboflavin/g. and 10–19 μg. of nicotinic acid/g.[12] This, together with its higher tryptophan content,[11] makes kaffircorn a useful addition to the high-maize diet which is common among the Bantu, particularly in the rural areas.

The changes in the vitamin content of the grain on malting are shown in Table IV. In the production of malt for brewing the sprouted grain is dried either in the sun or at temperatures below 50° ; ' kilning ' in the sense used by the barley maltsters is not practised. Malt produced under these conditions has a riboflavin content 40–70% higher than that of the original grain, while the nicotinic acid and thiamine contents are about the same. Kaffircorn malt is also used

in the preparation of some breakfast foods, for which purpose it is ' cured ' by subjecting it to much higher temperatures. This treatment causes a substantial loss of thiamine, but does not affect the other two vitamins. The breakfast foods prepared from kaffircorn consist essentially of a mixture of malt and grain flour, with some additional bran. The analysis of one such preparation is shown in Table III.

Table IV

Changes in vitamin content of kaffircorn during malting and curing

		Thiamine	Riboflavin	Nicotinic acid
		μg./g. dry basis		
Series I	Grain	5·5	1·2	40
	Malt, dried only	5·2	2·0	37
	Malt, cured	2·5	2·2	38
Series II	Grain	6·2	1·6	40
	Malt, dried only	8·5	2·2	37
	Malt, cured	3·5	2·4	40

Composition of kaffir beer

Since the bulk of the kaffircorn produced in South Africa is consumed in the form of beer, the nutritional value of this product and the changes which occur during brewing are of particular interest. Table V summarises the analyses of 21 samples of beer collected between 1954 and 1960 from nine municipal breweries which are amongst the largest and most modern in the industry. During the past 10 years there have been important changes in the proportions of raw materials used in brewing kaffir beer. As malts of improved diastatic power have become available, there has been a steady decrease in the ratio of malt to adjuncts used. In addition many breweries have changed over, partly or wholly, from the use of unmalted kaffircorn meal as adjunct to maize meal or grits. These changes have not yet become stabilised and there are still considerable differences in the raw materials used in different breweries. These are to some extent reflected in the differences in the composition of the beers examined.

Table V

Composition of municipal kaffir beers

	Range	Mean	No. of analyses
pH	3·2–3·7	3·4	10
Alcohol (% w/v)	1·8–3·9	3·0	17
Solids (% w/v)			
Total	3·0–8·0	5·4	17
Insoluble	2·3–6·1	3·7	6
Nitrogen (% w/v)			
Total	0·059–0·137	0·093	16
Soluble	0·010–0·017	0·014	9
Thiamine (μg./100 ml.)	20–230	93	21
Riboflavin (μg./100 ml.)	27–170	56	21
Nicotinic acid (μg./100 ml.)	130–660	315	21
Ascorbic acid (mg./100 ml.)	0·01–0·15	0·04	7

The values obtained in the present study for the ascorbic acid content of the beers are much lower than those found by Levy & Fox[13a] and Fox & Stone[13b] who reported 0·2–1·2 mg. ascorbic acid per 100 ml. These authors, however, found considerable interference due to extraneous reducing substances in their determinations by the indophenol method, and although they attempted to eliminate this by working between pH 1 and 2, it is possible that the values they obtained were still too high. This is supported by the fact that guinea-pig experiments showed kaffir beer to be practically devoid of antiscorbutic activity.[13b] In spite of this it is frequently stated that kaffir beer in the quantities consumed, provides a useful amount of ascorbic acid in the diet of the Bantu. According to our figures, however, it would be necessary to consume on an average 17 gallons of beer a day to provide the minimum requirement for an adult of 30 mg. of ascorbic acid. This is far in excess of the normal consumption of 2–8 pints.

The amount of thiamine, riboflavin and nicotinic acid in the beers examined varied considerably, some beers being very rich in these vitamins. Even at the lower levels, half a gallon of beer, which is the amount commonly consumed daily, supplies a substantial part of the adult requirement of B vitamins (Table VI). According to Golberg & Thorp[4a] African cereals usually provide adequate amounts of thiamine, but riboflavin and nicotinic acid deficiencies are widespread. The vitamins supplied by fermented foods such as kaffir beer are therefore of great importance.

Table VI

Amounts of B vitamins supplied by ½ gallon of kaffir beer
(calculated from values in Table V)

	Thiamine, mg.	Riboflavin, mg.	Nicotinic acid, mg.
Minimum	0·45	0·61	2·9
Maximum	5·2	3·9	15·0
Mean	2·1	1·3	7·2
Minimum daily requirement*	1·0	1·2	10·0

* United States Food & Drugs Administration[14]

Comparison of our figures with the earlier values of Golberg & Thorp[4b] is made difficult by the fact that they gave no indication of the range of values they encountered. Their mean values for the vitamin content of five municipal beers and five native beers were thiamine 57 and 36 μg., riboflavin 51 and 56 μg. and nicotinic acid 480 and 450 μg., respectively, per 100 ml. beer. It thus appears that, *on the average*, the vitamin content of municipal beers has changed little since 1945–46 and that it is similar to that of the home-brewed product.

Factors influencing the vitamin content of kaffir beer

To try to find the reason for the differences in the vitamin content of beers produced by different breweries, the changes in thiamine, riboflavin and nicotinic acid at different stages of the brewing process were followed in two brews made at different breweries, both brews made from kaffircorn malt and maize grits. The results are shown in Table VII.

The lactic acid fermentations were carried out somewhat differently. In the first brewery 100 gal. of a starter culture of *Lactobacillus delbrückii* containing malt 180 lb. and maize grits 45 lb. were added to malt 180 lb. in 200 gal. of water at 50°, and souring was allowed to proceed for 6 h. The whole of the sour was carried forward to the next stage. In the second brewery an inoculum consisting of 10% of sour from the previous brew was added to 180 lb. of malt in 400 gal. of water and souring was continued for 14 h. Of the product, 10% was retained as inoculum for the next brew and the remainder transferred to the mash aggregate. The latter procedure is the one used in most large breweries ; that used in the first brewery is an improved process designed to speed up the lactic fermentation. The rest of the process was the same in both breweries. Ten bags of grits (1800 lb.) and approximately 1500 gal. of water were added to the sour and the whole boiled for 2 h. at atmospheric pressure (94–95° in the Pretoria–Witwatersrand area). The liquid was cooled to 60°, the balance of the malt was added and the mash held at 60° for 2 h. before being cooled to 30–35°. The ' mash ' samples were taken at this stage. The dried culture yeast, dispersed in about 5 gal. of water, was added and straining of the mixture through coarse sieves was started immediately. The beers were sampled and sold after 48 h. fermentation at 30°.

The difficulties in sampling, particularly in the case of the soured porridge from the lactic acid fermentor and the spent grain, make it necessary to interpret results with some caution. There was no significant gain or loss of B vitamins in the lactic acid fermentation in the first brew or in thiamine and nicotinic acid in the second. In the latter case, however, there was a 200% increase in riboflavin content. To what extent this is due to differences in the souring processes used remains to be determined. In a similar study of a beer produced in the early days of the industry where souring was carried out in wooden vessels, without temperature control, over a period of 36 h., Golberg & Thorp[4b] found increases of 94, 56 and 13% in thiamine, riboflavin and nicotinic acid content, respectively. The considerable loss of thiamine during

Table VII

Changes in quantities of B vitamins during brewing of kaffir beer

Stage of brewing	Quantity of material	Vitamin content (μg./g. wet wt.)			Total vitamin content (mg.)			Increase (+) or decrease (−) %		
		Thiamine	Riboflavin	Nicotinic acid	Thiamine	Riboflavin	Nicotinic acid	Thiamine	Riboflavin	Nicotinic acid
Brew 1										
Lactic acid fermentation										
Raw materials										
Kaffircorn malt	360 lb.	4·70	3·10	39·5	770	510	6460			
Maize grits	45 lb.	2·50	0·36	6·6	50	8	140			
					820	518	6600			
Product										
Sour	300 gal.	0·64	0·29	4·6	870	400	6260	+6	−23	−5
Mashing										
Raw materials										
Sour	300 gal.	0·64	0·29	4·6	870	400	6260			
Maize grits	1800 lb.	2·5	0·36	6·9	2040	290	5650			
Kaffircorn malt	540 lb.	4·7	3·1	39·5	1150	760	9690			
					4060	1450	21,600			
Product										
Mash	1670 gal.	0·49	0·18	4·0	3720	1370	30,300	−8	−5	+40
Alcoholic fermentation										
Raw materials										
Mash	1670 gal.	0·49	0·18	4·0	3720	1370	30,300			
Yeast	5 lb.	46	29	355	105	66	760			
					3825	1436	31,060			
Products										
Beer	1550 gal.	0·20	0·27	1·7	1410	1900	11,900			
Spent grain	1310 lb.	0·33	0·53	7·9	200	320	4700			
					1610	2220	16,600	−58	+54	−47
Overall gain or loss on vitamins in raw materials (%)								−60	+43	−30
Brew 2										
Lactic acid fermentation										
Raw materials										
Kaffircorn malt	180 lb.	5·4	2·7	43	440	220	3500			
Product										
Sour	450 gal.	0·28	0·33	1·2	570	675	2450	+30	+207	−30
Mashing										
Raw materials										
Sour	450 gal.	0·28	0·33	1·2	570	675	2450			
Maize grits	1800 lb.	3·4	0·19	3·9	2780	155	3180			
Kaffircorn malt	760 lb.	5·4	2·7	43	1860	920	14,800			
					5210	1750	20,430			
Product										
Mash	1700 gal.	0·30	0·19	2·4	2320	1470	18,500	−55	−16	−9
Alcoholic fermentation										
Raw materials										
Mash	1700 gal.	0·30	0·19	2·4	2320	1470	18,500			
Yeast	5 lb.	34	32	248	76	73	563			
					2396	1543	19,063			
Products										
Beer	1600 gal.	0·27	0·28	2·3	1960	2040	16,700			
Foam	8 gal.	23	5·4	27	835	196	980			
Spent grain	880 lb.	0·50	0·50	7·4	200	200	2960			
					2995	2436	20,640	+25	+58	+8
Overall gain or loss based on vitamins in raw materials (%)								−42	+78	−6

mashing in the second brew agrees with the findings of the earlier workers. In the first brew, on the other hand, mashing proved less destructive to this vitamin.

The finding that there was a loss of thiamine and nicotinic acid during the alcoholic fermentation in the first brew studied was surprising at first sight. As in most breweries, however, the yeast used was a vigorous top fermenter and a certain amount of foaming over of the beer in the fermentor occurred at the height of the fermentation. In the second brew an attempt was made to collect this foam and estimate the amount of yeast and vitamins lost in this way. The figure of 8 gal. given in Table VII for the volume is minimal since it took into account only the collapsed foam collected from the overflow from the fermentor; the dried material adhering to the inside top of the vessel was not measured. As can be seen from the table, the foam was extremely rich in B vitamins, particularly in thiamine, so that the losses observed during the first fermentation could be explained on this basis.

Foaming can be readily controlled by breaking the foam just above the liquid surface by stirrers, and foam controllers have been installed on fermentors in some breweries. Table VIII showed the effect of foam control on the vitamin balance during the alcoholic fermentation. The two brews, numbers 3 and 4, were prepared at brewery No. 2 as described above, and were comparable in all respects except that one fermentor was fitted with a foam controller

Table VIII

Effect of foam control on vitamin balance during fermentation of kaffir beer

Stage of brewing	Quantity of material	Vitamin content (μg./g. wet wt.)			Total vitamin content (mg.)			Increase (+) or decrease (−) %		
		Thiamine	Ribo-flavin	Nicotinic acid	Thiamine	Ribo-flavin	Nicotinic acid	Thiamine	Ribo-flavin	Nicotinic acid
Brew 3 (no foam control)										
Raw materials										
Mash	1760 gal.	0·31	0·08	2·6	2480	640	20,800			
Products										
Beer	1570 gal.	0·20	0·36	2·0	1430	2560	14,300			
Foam	16 gal.	16	4·8	28	1160	348	2030			
Spent grain	1215 lb.	0·50	0·51	8·5	276	282	4680			
					2866	3190	21,010	+16	+398	+1
Brew 4 (with foam control)										
Raw materials										
Mash	1700 gal.	0·35	0·27	2·9	2700	2080	22,400			
Products										
Beer	1580 gal.	0·44	0·44	2·6	3160	3160	18,600			
Spent grain	1260 lb.	0·72	0·64	11	412	366	6300			
					3572	3526	24,900	+32	+70	+11

and the other not. In this case all the foam was collected in order to get an accurate assessment of the vitamins in this fraction. The results show that, where there was no control, approximately 40% of the thiamine and 10% each of the nicotinic acid and riboflavin in the fermentation mixture were lost in the foam. Where the yeast was retained in the mixture, as in brew 4, the beer had twice the thiamine content of the ordinary beer, as well as substantially higher contents of riboflavin and nicotinic acid. The extent to which loss of yeast in foam occurs during fermentation is therefore one of the factors which influences the vitamin content of the beer.

When allowance was made for the vitamins present in the foam, the present work showed that there was no significant synthesis of nicotinic acid during the alcoholic fermentation, while there was a small increase (16–32%) in thiamine and a substantial increase (70–80%) in riboflavin. (The riboflavin content of the mash in brew 3 was abnormally low, so that the results for this brew have not been taken into consideration here.) Golberg & Thorp,[40] on the other hand, reported increases of 38, 141 and 21% for nicotinic acid, thiamine and riboflavin,

respectively, during the fermentation of their beer. They used no culture yeast and the fermentation was brought about by the wild yeasts on the malt. It is probable therefore that the vitamin production of the yeasts present in their fermentation was quite different from those used in the brews studied here.

Another factor which may influence the vitamin content of the beer is the nature of the raw materials used. As can be seen from Tables III and VII, maize grits contain only about one-tenth as much nicotinic acid and riboflavin as does kaffircorn grain. From this point of view, the present tendency to use the former as an adjunct instead of the latter is unfortunate, particularly from the point of view of the nicotinic acid content of the beer, since there is little or no synthesis of this factor during brewing. It is probably significant that the five beers which were found in the present study to have the highest nicotinic acid (>500 μg./100 ml.) were brewed either solely from kaffircorn malt and grain or with less than 10% of maize products. The preference for maize grits is based partly on price and partly on the fact that it gives a beer with better body and texture than does kaffircorn grain. It may be necessary to consider whether these factors are not outweighed by the lowered nutritive value of the beer.

Conclusions

Kaffir beer has always been regarded not merely as an alcoholic beverage but also as a food for the Bantu. The present study confirms the earlier findings that it is of considerable value as a source of B vitamins in the diet. It has also shown, however, that the raw materials and methods employed in its manufacture may influence the vitamin content considerably. Any attempts to introduce improved methods of brewing should therefore take into consideration the effect these may have on the nutritive value of the beer.

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South African Council for Scientific & Industrial Research
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KAFFIRCORN MALTING AND BREWING STUDIES. IX.*— Amino-Acid Composition of Kaffircorn Grain and Malt

By P. J. HORN and H. M. SCHWARTZ

The amino-acid compositions of three varieties of kaffircorn and of one sample of kaffircorn malt were determined. Proline was present in relatively large amounts. From the point of view of human nutrition, lysine was deficient, while methionine was the second limiting amino-acid.

Introduction

The amino-acid compositions of three varieties of grain sorghum or kaffircorn (*Sorghum vulgare*) from South Africa and of one sample of kaffircorn malt have been determined as part of a wider study of the nutritive value of kaffircorn products.[1]

The essential amino-acids in sorghum grown in different parts of the world have been determined by various workers. Adda[2] and Adrian[3] analysed a number of peasant-cultivated and experimentally grown species from the Senegal, Baptist[4] reported values for one sample from Ceylon, while in India Balasubramanian[5] and Chitre[6] and their co-workers examined various strains of 'cholam' and 'jowar' (*Sorghum vulgare*). Finally Williams,[7] Vavich *et al.*[8] and Lyman *et al.*[9] have analysed sorghums from the United States. When the present studies were undertaken there were no figures available for the complete amino-acid analysis of sorghum grain, nor any analyses of sorghum malt, in spite of the fact that much of the cereal is consumed in Africa and Asia in the malted state. Recently, however, complete analyses of the amino-acids in sorghum grain and malt from the Belgian Congo have been published by Close & Naves.[10]

Experimental

Materials

Samples of grain of the short red, white and birdproof varieties of kaffircorn were obtained from the Potchefstroom Agricultural College. These represent three of the most important varieties of kaffircorn grown in South Africa.

The malt was a commercial sample prepared from short red kaffircorn. It had a diastatic value of 30·1 K.D.U./g.[11]

All samples were milled to pass a 60-mesh sieve.

Estimation of the amino-acids

Tryptophan, cysteine + cystine and methionine were determined microbiologically by the methods described by Barton-Wright.[12] The remaining amino-acids were determined chromatographically according to the technique of Moore & Stein.[13] Approximately 600 mg. of sample were hydrolysed by refluxing for 24 h. with 200 ml. of redistilled 6N-HCl.[14] Determinations were carried out at least in duplicate, separate hydrolysates being prepared for each chromatogram. No correction was applied for destruction of amino-acids during hydrolysis.

Results and discussion

The results are given in Table I.

Of the total nitrogen in the samples, 78–86% was present as free and combined amino-acids. The balance is probably accounted for by the presence of other nitrogenous compounds in the grain and malt rather than by loss of amino-acids during hydrolysis, since it was shown that the procedure employed causes negligible destruction of amino-acids, even in the presence of large amounts of carbohydrate.[15] In this connexion, it is of interest that Close & Naves[10] found that glucosamine was present to the extent of approximately 0·5% of the total nitrogen in the samples they examined.

The proportions of the amino-acids in the three varieties of kaffircorn and in the malt were very similar. There is thus little or no change in the total quantities of the amino-acids in the grain during malting, although there is a considerable change in the amounts present in

* Part VIII: preceding paper

Table I

Amino-acid composition of kaffircorn grains and malt

	Amino-acid, g./100 g. of grain or malt				Amino-acid, g./100 g. of crude protein (N × 6·25)			
	Short red grain (1·36% N)	White grain (1·79% N)	Birdproof grain (1·63% N)	Short red malt (1·47% N)	Short red grain	White grain	Birdproof grain	Short red malt
Aspartic acid	0·623	0·953	0·783	0·857	7·33	8·52	7·69	9·31
Threonine	0·288	0·378	0·333	0·318	3·39	3·38	3·27	3·46
Serine	0·356	0·473	0·458	0·393	4·19	4·23	4·50	4·27
Glutamic acid	1·54	2·31	1·99	1·62	18·1	20·6	19·5	17·6
Proline	0·697	0·945	0·826	0·775	8·20	8·45	8·11	8·42
Glycine	0·280	0·334	0·308	0·285	3·29	2·99	3·02	3·10
Alanine	0·717	1·08	0·919	0·738	8·44	9·65	9·02	8·02
Valine	0·386	0·608	0·525	0·436	4·54	5·43	5·15	4·74
Methionine	0·174	0·137	0·176	0·122	2·05	1·22	1·73	1·33
Isoleucine	0·308	0·504	0·429	0·348	3·62	4·51	4·21	3·78
Leucine	0·975	1·54	1·30	0·989	11·5	13·7	12·7	10·8
Tyrosine	0·358	0·492	0·461	0·368	4·21	4·40	4·53	4·00
Phenylalanine	0·407	0·581	0·511	0·440	4·79	5·19	5·02	4·78
Lysine	0·210	0·241	0·244	0·259	2·47	2·15	2·40	2·81
Histidine	0·208	0·274	0·242	0·226	2·45	2·45	2·38	2·46
Arginine	0·477	0·721	0·611	0·460	5·61	6·44	6·00	5·00
Cystine/2	0·101	0·103	0·152	0·111	1·19	0·92	1·49	1·21
Tryptophan	0·154	0·138	0·123	0·099	1·81	1·23	1·21	1·08
Amino-acid N as % of total N					80·0	86·4	83·5	78·4

the free and combined states.[16] An interesting feature of the results is the high proportion of proline present. This reflects the high content of prolamines which comprise 46–60% of the proteins in the malt and grain.[16]

The values obtained in the present study are in close agreement with those of Close & Naves[10] except in the case of arginine where our figures (5·0–6·4 g./100 g. of crude protein) are somewhat higher than theirs (3·8–3·9 g./100 g.). Most other workers have also reported lower values, ranging from 2·9 to 4·4 g./100 g. for this amino-acid, an exception being Chitre and his co-workers[6] whose figures (5·6–6·5 g./100 g.) are of the same order as ours. In general the values for the essential amino-acid contents of sorghums reported from different parts of the world are very similar. One exception is the value of 24·1–25·2 g./100 g. of crude protein for leucine reported by Adrian[3] which is very much higher than the values of 8·9–15·4 g./100 g. found by all other workers including ourselves.

From the point of view of human nutrition, kaffircorn is deficient in lysine which is present to the extent of only about half the estimated requirement for infants and adults, viz., 3·2–5·6 g./100 g. of protein.[17] The next limiting amino-acid would appear to be methionine, the content of which approaches the minimal requirements of 0·9–1·5 g./100 g. protein.[17] While the content of tryptophan is not very much greater than the estimated requirement, it is considerably higher than that in maize, so that consumption of kaffircorn grain or malt helps to make good the deficiency of this amino-acid in a diet in which maize is the staple food, as in the case of the Bantu of South Africa.

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National Chemical Research Laboratory
South African Council for Scientific & Industrial Research
Pretoria

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STUDIES ON EGG SHELLS. XVI.*—Variations in Shell Thickness over Different Parts of the same Shell

By C. TYLER

Variations in shell thickness over the surface of the egg have been studied in a number of normal and abnormally thin shells. There is far less variation around the latitudes of the egg than there is longitudinally. Different birds give, for normal shells, different patterns of thickness from pole to pole, but for any one bird the pattern is usually fairly constant. The most frequent general pattern is one in which the broad and narrow caps each have a thickness greater than most intermediate latitudes, but the minimum value may fall in different positions in different eggs.

Abnormally thin shells seem to deviate from the normal pattern for the same bird, but it will be necessary to carry out more work on this before reliable conclusions can be drawn.

With a collection of eggs, the thickness of one latitude is highly correlated with the thickness of another latitude and it is possible to calculate from assumed values for one latitude the pattern for the rest of the shell.

Various methods for obtaining an accurate value for mean shell thickness by using a sample of measurements have been compared.

Introduction

A method of dismantling an egg shell whereby variations over the surface of the shell may be studied in relation to a number of characteristics has been described by Tyler.[1] The present paper is concerned with studies on the variations in shell thickness in normal shells and abnormally thin shells.

Experimental

Material

The 52 normal shells studied came from the eggs from 15 birds of various breeds, ranging from single eggs up to a clutch of 8. In some cases a number of clutches was available from the same bird. The shells varied in mean thickness from 241 to 371 μ and, therefore, covered a very wide range. One bird, in addition to laying a few normally shelled eggs, produced a succession of twelve thin-shelled eggs and these were also studied, as well as six thin-shelled eggs from a variety of other birds.

Methods

The method of dismantling the shell and making the measurements was fully described in an earlier paper.[1] It is sufficient to state here that the shell is divided into six latitudinal

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collars, each 1 cm. in width, and two caps one from each pole. The latitudinal collars are then each sub-divided longitudinally into eight pieces, which for any one collar are of equal length. The pieces are marked with a letter indicating the collar and a number indicating the position on the collar. A series of six pieces lettered B–G and carrying the same number will, therefore, represent a longitudinal segment, which is completed by an appropriately numbered segment from each cap.

Since all collars are measured from the equator of the egg and since the collars are always 1 cm. wide, it follows that variations in egg size will decide the size of the two caps. With small caps only one set of latitudinal readings has been made around the caps, with larger caps it has been possible to make two sets of readings. This is not a major difference in technique and only affects the total number of readings made per cap.

In the earlier paper measurements made in two parallel rows on each piece were averaged to give the mean thickness for that piece. The same type of measurement was made in the present study but the values have been arranged differently. A row of three measurements on each piece taken from one collar, in actual fact, represents a total of 24 measurements, all made very close to one latitude. Therefore, the variation between measurements on one latitude may be studied and if the measurements for all other latitudes are similarly taken, then the mean values for each latitude may be compared. One or two sets of latitude values are also available for each cap. This method of calculation has been used in this paper. For some purposes, as will be explained later, the mean value of the two latitude means for one collar has been used.

With normal eggs the shell thicknesses have been measured with an anvil-jawed micrometer screw gauge. For the thin-shelled eggs discs of shell of equal size were cut with a cork borer and the calcium dissolved in acid and determined analytically. Multiplication by the appropriate factor then gave mg. of Ca per cm.^{2} Variations in the absolute quantity of Ca per cm.^{2} therefore represent a measure of variation over the surface of the shell.[1]

When shell thicknesses are being measured accurately, it is necessary to remove the shell membrane first and to do this efficiently the pieces of shell must be boiled in 2·5% sodium hydroxide solution.[2] This means that the membrane is destroyed and similar studies on the shell membrane must normally be carried out on other eggs.

From the results in the previous paper and from many more results in this paper, it is clear that latitudinal variations are very small. Therefore, by taking four alternate segments for thickness studies and the other four alternate segments for membrane studies, it is possible to obtain data for both shell thickness and membrane thickness on the same shell. The membrane values will be discussed in another paper but the point is mentioned here because values for shell thickness for eggs from birds 500, 502 and 503 are derived from measurements on four segments instead of the full eight.

Results

Latitudinal and longitudinal variations

It was pointed out in the earlier paper[1] that the eight pieces of one collar tended to give very similar results for thickness but that there were considerable differences between collars, i.e., the variation in thickness of the shell was almost entirely owing to variations from pole to pole and not to variations around one collar.

This has been amply confirmed in this study. The results from a total of ten eggs from a selection of three birds were calculated in the form of mean thickness per piece and the coefficient of variation (C.V.) then calculated in relation to the eight pieces in a collar. All six collars were considered for each egg but the two caps were ignored since readings for the caps could not be treated in the same way.

The results are set out in Table I which shows quite clearly that the C.V. are very small for any particular collar, and the highest reading of all is only 1·78%. There is some indication that the variation within a collar may vary in relation to the position of that collar and the mean values for each bird show that the variation in general tends to be higher near the poles than elsewhere.

Table I

Shell thickness (μ): coefficients of variation between the eight values of any one latitudinal collar

Ten normal eggs from the three birds G, H and 124
Four thin-shelled eggs from bird L (Ca, mg./cm²)

Collar	Bird G Eggs					Bird H Eggs			
	1/1	1/2	1/3	1/4	Mean	1/1	1/2	1/3	Mean
B	1·05	1·18	0·92	1·56	1·18	0·75	0·95	0·97	0·89
C	0·49	0·55	0·69	0·65	0·60	0·37	0·75	0·31	0·48
D	0·51	0·66	0·82	0·98	0·74	0·34	0·50	0·64	0·49
E	0·72	0·71	0·82	0·86	0·78	0·58	0·45	0·34	0·46
F	0·24	0·49	1·08	0·83	0·66	0·37	0·60	0·31	0·43
G	1·78	0·76	0·88	1·65	1·27	0·58	0·73	1·28	0·86
Mean	0·80	0·73	0·87	1·09	0·87	0·50	0·66	0·64	0·60

Collar	Bird 124 Eggs				Bird L Eggs			
	1/1	1/2	1/3	Mean	1	2	7	10
B	0·63	0·76	0·51	0·63	9·93	10·09	1·38	5·23
C	0·43	0·45	0·42	0·43	13·83	22·82	4·18	2·78
D	0·68	0·23	0·35	0·42	21·03	33·20	3·22	0·54
E	1·14	0·36	0·36	0·62	13·43	19·01	0·65	0·82
F	1·14	0·60	0·74	0·83	9·43	14·07	1·56	3·45
G	0·47	1·23	0·66	0·79	14·51	8·82	0·73	1·29
Mean	0·75	0·61	0·51	0·62	13·69	18·00	1·95	2·35

In the eggs from bird L, the Ca, mg./cm.² (a measure of thickness), was 1·20, 1·07, 12·3 and 17·6 for 1, 2, 7 and 10 respectively.

It will also be observed that there may be a greater variation in all collars for one bird compared with all collars for another bird. Thus bird G shows a greater mean C.V. than birds H or 124.

The results for four of the thin-shelled eggs have been included in Table I and it is immediately evident that thin-shelled eggs show considerable variation within one collar, between different collars of the same egg and between different eggs. The values range from 0·54 up to 33·20%, so that clearly in the early stages of calcification there is far greater variation even within collars than there is later. The mean values for the four eggs also show that the very thin-shelled eggs, L/1 and L/2, are more variable than the less thin shells of eggs L/7 and L/10.

Table II

Shell thickness (μ): variation between measurements made along 12 lines of latitude (collars) and along 24 lines of longitude (segments) (Egg X/1)

Analysis of variance

	Degrees of freedom	Sums of squares	Variance	F
Collars	11	68,598	6236·18	418·54***
Segments	23	392	17·04	1·14 N.S.
Error	253	3769	14·90	
Total	287	72,759		

An even closer study was made of one egg, namely, X/1, by taking the total of 288 measurements made on all the pieces except the two caps. These measurements gave a pattern of 12 lines of latitude each with 24 readings or 24 lines of longitude each with 12 readings. An analysis of variance was carried out on the values arranged in this way. Clearly the variations (Table II) between lines of latitudes, i.e., along longitudes, are very large compared with the error and the variations between lines of longitudes, i.e., along latitudes, is no greater than the error. This confirms the previous results and emphasises the smallness of the variation around a line of latitude. The pattern of changing thickness in normal eggs is therefore almost entirely limited to variations along the lines of longitude from pole to pole. Fig. 1 shows the variation

around the latitude giving the lowest mean thickness and that giving the highest mean thickness for eggs X/1 and I/1. This serves to underline the very great differences between latitudes compared with the small variations within a latitude. Because of this, the remainder of the results will be considered in terms of mean values for each line of latitude which will give the pattern of variation from pole to pole, or in terms of mean values for each pair of lines of latitude taken from one collar.

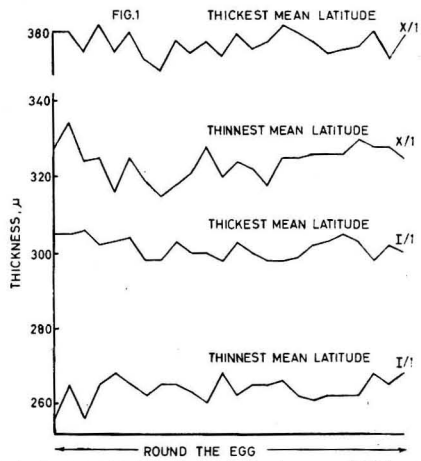


FIG. 1.—*Latitudinal variations for the latitudes with the lowest and highest mean shell thickness* (μ) Eggs X/1 and I/1

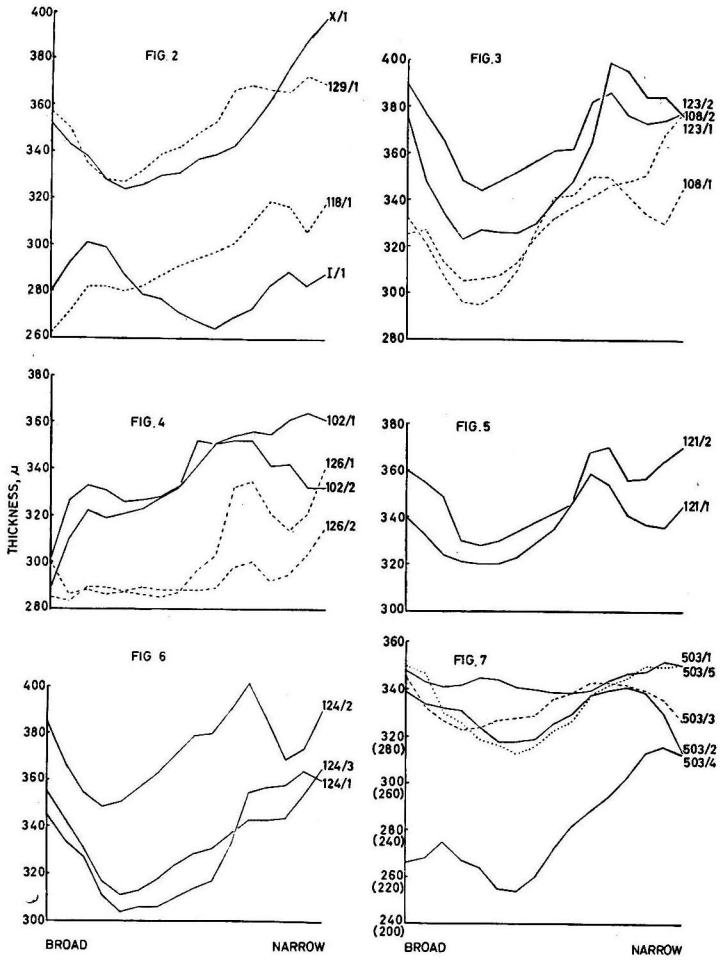
Patterns of thickness for normal shells

The results are set out in Figs. 2–13. Fig. 2 shows the results for four eggs taken from four separate birds and exhibits four patterns which are quite different. The shell thickness of egg 118/1 shows a fairly steady increase from the broad pole A to the narrow pole H, whilst I/1 increases, then decreases, then increases again. The other two, X/1 and 129/1, each give similar patterns from the broad end to past the waist, then they differ considerably. In Figs. 3–5, patterns for two-egg clutches are shown for five birds. Again, different birds give different patterns, but, generally speaking, the two eggs from one clutch are fairly similar. A three-egg clutch was obtained from bird 124 (Fig. 6), and two of these gave very similar patterns but the third was different. However, this difference was chiefly associated with a thickening of the shell in a band at one particular point.

Bird 503 (Fig. 7) gave a five-egg clutch and there were no great differences in pattern, except for egg 503/4 which was entirely different, and bird 502 (Fig. 8) yielded an eight-egg clutch in which seven eggs were surprisingly similar in pattern, except for one measurement, but egg 502/4 was very different.

Two separate two-egg clutches were obtained from bird 500 and all these four eggs were very similar in pattern (Fig. 9) and again, bird G gave two four-egg clutches separated by a single egg. It will be seen that the general patterns of most of these were very much alike (Figs. 10 and 11). Finally, three successive three-egg clutches from bird H (Figs. 12 and 13) gave patterns which were exceedingly variable from egg to egg.

Despite the vastly different patterns, particularly between birds, it is nevertheless clear that, in the majority of cases, the caps of the egg shell are thicker than the remainder, but that the thinnest part is not always in the same latitude. Sometimes it is nearer to the broad pole, sometimes nearer to the narrow pole, and sometimes at the equator.



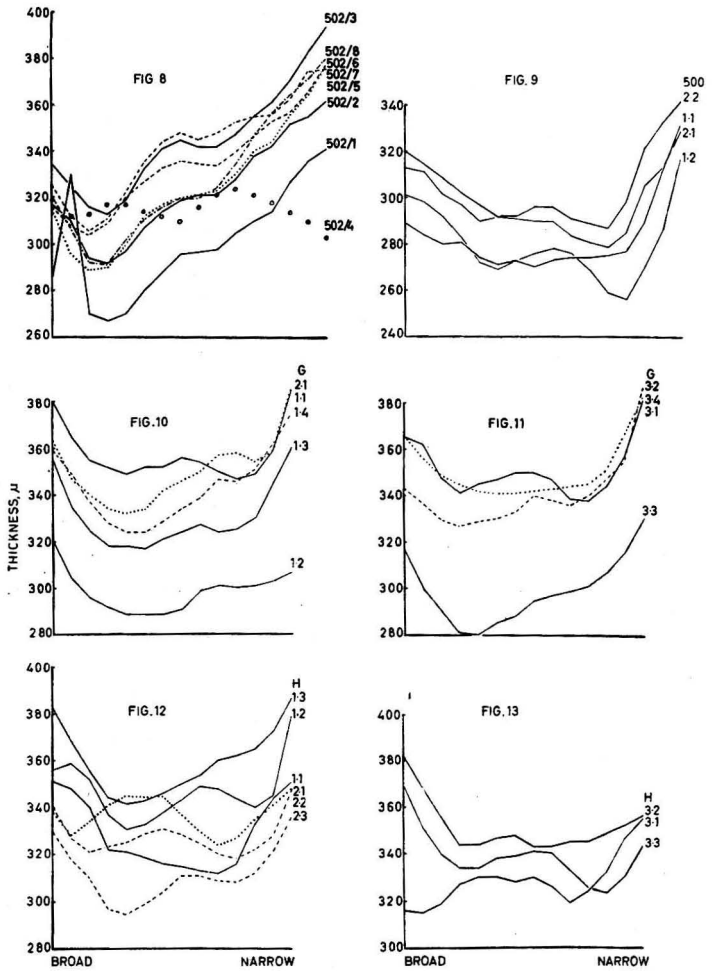
FIGS. 2–7.—*Variations in shell thickness (μ) from the broad to the narrow pole of normally shelled eggs*

Single digits after the bird number indicates eggs from only one clutch. Two digits indicate two or more clutches, e.g., 2.3 is the third egg of the second clutch. Bracketed values on the scale of Fig. 7 refer only to egg 503/4.

Patterns of thickness for thin shells

It must be borne in mind that the measurements on the thin shells were made in an entirely different manner and are expressed as mg. of Ca/cm.^{2} For purposes of comparison, it can, however, be stated that, very approximately, a piece of normal shell, say 300 μ thick, would give about 250 mg. of Ca/cm.^{2} Thus the shells under consideration are very thin.

In most of the thin-shells of bird L (Fig. 14), it would appear that the pattern is roughly similar to the general one for normal eggs, namely, both caps thicker than the intervening shell, but again the thinnest portion is not always in the same relative position. However, when



FIGS. 8–13.—*Variations in shell thickness* (μ) *from the broad to the narrow pole of normally shelled eggs*

Single digits after the bird number indicate eggs from only one clutch. Two digits indicate two or more clutches, e.g., 2.3 is the third egg of the second clutch. Figs. 8 and 9 have 16 readings from pole to pole, while Figs. 10–13 each have only 14 readings.

the normal shells from this same bird are examined (Fig. 15), it is seen that the pattern is quite different, for here there is a thicker ridge below the equator and the narrow pole is usually the thinnest part of the shell. It is impossible to predict how the thin shells would have developed if development had not been stopped, but there is no doubt that the pattern for the normal shells of this bird is entirely different from the pattern for the thin shells. Fig. 16 shows the patterns for a selection of other thin-shelled eggs and it is evident that there can be a considerable degree of variation.

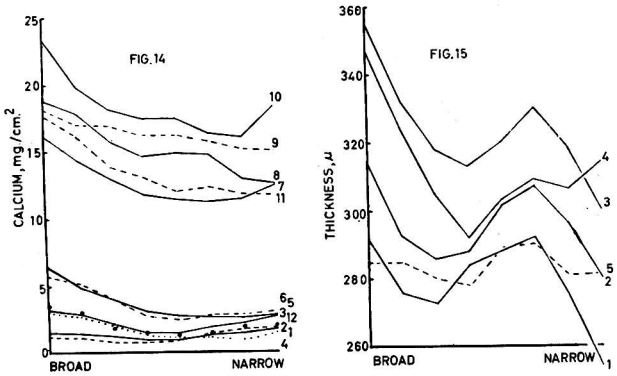


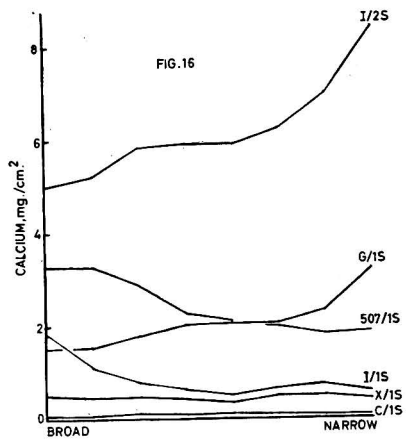
FIG. 14.—*Variations in shell thickness (Ca, mg./cm.²) from the broad to the narrow pole of abnormally thin-shelled eggs from bird* L

Eggs in succession but clutches not known

FIG. 15.—*Variations in shell thickness (μ) from the broad to the narrow pole of normally shelled eggs from bird* L

Eggs laid shortly before those in Fig. 14, but order not known

FIG. 16.—*Variations in shell thickness (Ca, mg./cm.²) from the broad to the narrow pole of abnormally thin-shelled eggs from different birds*



Discussion

General

There is every indication that individual birds over short periods of time tend to produce shells with a typical thickness pattern, but it must be admitted that occasionally an egg is produced which deviates widely from the rest. It is impossible to say at this stage whether this is owing to some major disturbance in shell deposition or not. Only one hen, namely, H, produced a collection of eggs without any obvious general pattern and this again cannot be accounted for at present.

The results with the thin-shelled eggs, at least suggest that the pattern at one stage of shell formation may not necessarily be the same at a later or earlier stage of shell formation, and this is a line which needs to be followed in more detail in future work.

Furthermore, it must not be forgotten that with thin-shelled eggs the variation within collars is also much larger than it is for normally shelled eggs.

Thickness relationships in different parts of the shell

In order to study this problem, the mean thicknesses of each of the two caps and six collars have been set down for each of the 52 normal eggs and a correlation coefficient calculated for the relationship between cap A and the adjacent collar B. Similar correlation coefficients have been calculated for all other combinations (see Table III). It will be seen that the readings are duplicated on each side of the diagonal but this is merely to make clearer the calculation of the column means. The values in any one column refer to the relationship of the cap or collar shown at the head of that column to the other caps and collars.

Table III

Correlation coefficients for the relationships between the thickness of shell in different caps and collars and of these to mean shell thickness (M)

Normal shells (52)

	A	B	C	D	E	F	G	H	M
A	—	0·946	0·868	0·830	0·791	0·686	0·696	0·716	0·859
B	0·946	—	0·921	0·848	0·809	0·681	0·672	0·655	0·863
C	0·868	0·921	—	0·970	0·917	0·796	0·796	0·746	0·952
D	0·830	0·848	0·970	—	0·964	0·854	0·851	0·803	0·974
E	0·791	0·809	0·917	0·964	—	0·933	0·887	0·794	0·974
F	0·686	0·681	0·796	0·854	0·933	—	0·939	0·766	0·922
G	0·697	0·672	0·796	0·851	0·887	0·939	—	0·883	0·923
H	0·716	0·655	0·746	0·803	0·794	0·766	0·883	—	0·852
Means of columns	0·791	0·790	0·859	0·874	0·871	0·808	0·818	0·766	0·915

(Column M is *not* the mean of the rows but the correlation coefficient between mean shell thickness and each individual cap and collar thickness)

Abnormally thin shells (12)

	A	B	C	D	E	F	G	H
A	—	0·998	0·993	0·990	0·986	0·986	0·985	0·983
B	0·998	—	0·997	0·994	0·990	0·991	0·988	0·978
C	0·993	0·997	—	0·999	0·997	0·997	0·995	0·986
D	0·990	0·994	0·999	—	0·999	0·999	0·998	0·987
E	0·986	0·990	0·997	0·999	—	0·999	0·998	0·987
F	0·986	0·991	0·997	0·999	0·999	—	0·998	0·984
G	0·985	0·988	0·995	0·998	0·998	0·998	—	0·992
H	0·983	0·978	0·986	0·987	0·987	0·984	0·992	—
Means of columns	0·989	0·991	0·995	0·995	0·994	0·993	0·993	0·985

It is important to make it quite clear at the outset, that, since the shells cover a wide range of thicknesses, and since any adjacent collars do not differ greatly in comparison with their total thickness, then high and significant values are to be expected for the correlation coefficients. Thus it is the trends of the correlation coefficients rather than their magnitudes or their significance, which are of importance. The values are directly comparable because all are based on 52 pairs of readings.

The first point to be noted is that, with few exceptions, for all collars the correlation coefficient between any two collars or between a collar and a cap, becomes smaller the further they are apart—a finding first noted by Olsson.[3] This is very well illustrated with collars D and E which lie immediately on each side of the equator of the egg. This finding is, of course, not surprising, but when cap H is considered, it will be seen that the rule does not hold. Collars E and F in close proximity to cap H give correlation coefficients lower than those for collar D which is further away. Cap A shows exactly the same relationship as the collars, namely, a decreasing correlation cofficient with increasing distance apart, except in its relationship to G which is slightly greater than to F and a larger increase again to H. It would thus appear that the cap H which is at the narrow pole, does not fit in with the relationships shown to each other by other parts of the shell.

It should also be noted that each cap and the six collars are highly correlated with mean thickness and that the two collars nearest the equator show the largest correlation coefficients, which decrease in magnitude the further the collar is from the equator. The two caps thus give the two lowest values. It is, however, important to point out that a series of measurements round the waist is not necessarily a satisfactory guide to thickness for comparative purposes.

A similar set of correlation coefficients is also given in Table III for the twelve thin-shelled eggs from bird L. All these are very close to unity but, again, when collars are considered, adjacent ones give a higher correlation coefficient than the ones further apart. Once more this is not evident with cap H which seems to be out of line with the rest of the shell. More work will have to be done to establish the physiological significance, if any, of this finding.

Table IV

Regression equations relating collar D to all other collars and caps (1) *for* 52 *normal shells and* (2) *for* 12 *abnormally thin shells*

Normal shells (μ)	Thin shells (Ca, mg./cm.^{2})
A = 0·942 D + 30·9	A = 1·191 D + 1·457
B = 0·852 D + 47·7	B = 1·079 D + 1·230
C = 0·924 D + 19·7	C = 1·025 D + 0·541
E = 0·968 D + 16·3	E = 0·995 D − 0·160
F = 0·956 D + 27·0	F = 0·948 D + 0·148
G = 0·872 D + 59·6	G = 0·889 D + 0·372
H = 0·853 D + 78·5	H = 0·923 D + 0·568

In addition to correlation coefficients, regression equations have also been calculated relating the thickness of the D collar to each of the other collars and caps; the choice of this collar being made because it is most highly correlated with other parts of the shell. These equations are given in Table IV and also values of each collar for assumed values of the D collar are shown in Fig. 17. It must be clearly understood that these curves represent ideal eggs of differing shell thickness and not any particular egg, but the curves suggest that, generally speaking, the increase in thickness is fairly uniformly spread over the whole shell, and that the pattern changes somewhat as shells become thicker. These results are promising but similar relationships calculated from sets of normal eggs from individual birds would be of far greater interest.

The twelve eggs from bird L are thus clearly of interest, even though they are abnormally thin. Table IV also shows the corresponding equations and Fig. 18 the curves calculated for

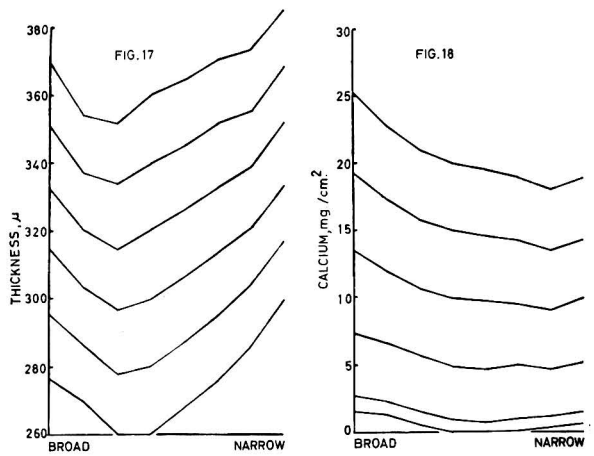


FIG. 17.—*Calculated patterns of shell thickness* (μ) *from the broad to the narrow pole for ' ideal ' eggs of different mean thickness*

FIG. 18.—*Calculated patterns of shell thickness* (*Ca, mg./cm.*^{2}) *from the broad to the narrow pole for abnormally thin-shelled eggs from bird L*

assumed values of the D collar. It would appear that calcification starts at the poles first and that even in the thinnest shell, the broad pole has slightly more calcium than the narrow pole. As calcification proceeds, the difference in favour of the broad pole becomes more and more pronounced. It must, however, be remembered that all these shells are still very thin so that no conclusions may be drawn as to what would happen with normal shells from this bird.

Measurement of shell thickness

The results obtained here are also useful in helping to decide how many measurements are required to obtain a reliable value for mean shell thickness and also whether particular points should be measured instead of taking points at random.

To test this, egg X/1, which showed considerable variation from pole to pole, was studied and ten readings from all the available 306 were taken at random. This was repeated for eight samples. Each of these eight samples were then bulked to give four samples of 20, then two of 40 and, finally, one of 80 readings. The values are given in Table V and may be compared with the mean for the 306 readings, namely, 342·2 μ which will be referred to as the true value. It will be seen that some samples, even one of 40 pieces, vary from the true value and that the large sample of 80 readings gives a result which is +6·3 μ in error.

Table V

Shell thickness (μ): *measurements based on random samples of ten measurements and combinations of these*

(Egg X/1)

Number of sample							
1	2	3	4	5	6	7	8
351·5	354·4	347·7	356·7	353·2	338·1	343·4	342·4
	353·0		352·2		345·7		342·9
		352·6			344·3		
			348·5				

True value based on 306 measurements = 342·2 μ

Since it is known that variations along lines of latitude are very small, it should be possible to get a reliable result by taking a set of measurements along a line of longitude. This has been done for egg X/1 for eight different lines of longitude with certain minor variations (see Table VI). For this particular egg, along any line of longitude, one measurement was available for each pole, one measurement from each of two caps, and two measurements from each of six collars. Using these 16 measurements (column *a*), it is seen that the individual lines of longitude give fairly consistent mean values, but that the maximum and minimum difference from the true value are +7·0 μ and +3·5 μ. This is caused by the extremely thick shell at each pole and, clearly, since these are only two isolated points on the whole surface, they should not carry so much weight, and it is best to ignore them. The fact that such measurements are very often anomalous when compared with the rest of the shell supports this. Taking the same measurements less the two poles, gives values (column *b*) which vary from the mean by a maximum of +3·2 μ and a minimum of +0·3 μ. These are an improvement on the previous method.

In addition to the poles being anomalous, it is true to say that the caps themselves tend to be thicker than the rest of the shell and since the cap is usually much smaller in area than the smallest collar, these, too, should not be allowed to carry too much weight. Therefore, the 12 results taken only from collars have been calculated (column *c*) and here the maximum error is −3·2 μ and the minimum −0·6 μ. There is thus little to choose between the second and third method and hence it would seem reasonable to use the one needing the smaller number of measurements, namely, 12 measurements spread out at intervals of 0·5 cm. along a line of longitude with the two centre ones 0·25 cm. on each side of the equator. This compares very favourably with the 80 random measurements which might or might not give a better value. Olsson[3] suggested five longitudinal measurements and from his diagram it has been possible

Table VI

Shell thickness (μ)*: evaluation of mean thickness by measurements made along eight different lines of longitude*

(Egg X/1)

Methods: (*a*) 16 measurements, including a single measurement for each pole
(*b*) 14 measurements, i.e., excluding the values for the poles
(*c*) 12 measurements, i.e., as (*b*) and, in addition, excluding the other measurements made on the caps
(*d*) 5 measurements, as in Olsson's method[3]

Longitude	Method			
	(*a*)	(*b*)	(*c*)	(*d*)
1	348·9	345·1	341·0	345·8
4	348·9	345·1	341·4	344·2
7	349·2	345·4	341·2	344·6
10	346·7	343·0	339·0	341·8
13	345·7	341·9	339·0	339·6
16	348·1	344·3	341·0	345·2
19	346·4	342·6	339·3	341·4
22	349·0	345·2	341·6	346·4

True value based on 306 measurements = 342·2 μ

to select the measurements made on egg X/1 which are nearest to the points where he made his measurements. In Table VI these values are given for the same eight longitudes (column *d*) and it will be seen that the maximum error is +4·2 μ and the minimum −0·4 μ.

None of the four methods set out in Table VI nor the random measurement of 80 pieces gives a really serious error. Methods using a series of longitudinal readings obviously have an advantage over the random measurements in that far fewer readings are required. Owing to the fact that the readings at the poles are often widely different from the rest of the shell, it seems reasonable to omit these.

Many workers have taken measurements round the waist. The mean value of 24 such measurements of egg X/1 was 333·9 μ which is 8·3 μ less than the true value. This is a larger error than most of the other methods give and from the evidence of the various patterns it is clear that measurements round the waist could be greatly in error as a method of assessing the mean shell thickness.

It may be argued that if a series of points along a line of longitude are to be taken, then, to obtain a really accurate result, the value for each point should be weighted by multiplying by the latitudinal circumference passing through that point before adding the values and taking the mean. This is true in theory, but in practice it can be shown to be unnecessary. If each collar is supposed to be devoid of longitudinal curvature, then it is easy to calculate the area of shell represented by each of the six collars. Thus, taking collar D as unity, the collars have approximately the relative areas shown in Table VII. The values for the two caps have been calculated on the assumption that the two caps were about average size. Using these factors in relation to the two caps and six collars of shell X/1, the mean value was 341 μ and the mean value without these factors was 342 μ. Other eggs gave similar results. Furthermore, using larger and smaller factors for the two caps which might vary in size from egg to egg made no more than a difference of about 2 μ in the final result. There is thus nothing to be gained from weighting the longitudinal readings according to collar size. This is explained by the fact that, although variation occurs along a longitude, it is usually relatively small when compared with total thickness.

Table VII

Approximate ratios of collar and cap areas (collar D as unity)

Cap or collar	Ratio	Cap or collar	Ratio
A	0·25	E	0·90
B	0·65	F	0·75
C	0·90	G	0·45
D	1·00	H	0·25

Thus, apart from measurements round the waist, all the methods normally used give very good results and there is no reason to suppose that tests on other eggs would reveal anything

different. A series of 6–12 measurements along a line of longitude, ignoring the poles, seems to combine accuracy with speed, if a mean value for shell thickness is required. This, however, is based on the assumption that for some reason the weight per unit area of true shell, as recommended by Tyler & Geake[4] is not to be used. If the pattern of variation from pole to pole is required, then readings taken at each pole plus about 12 equidistant readings along a line of longitude will be very satisfactory.

Conclusions

The results reported in this paper suggest that variations in shell thickness are worthy of closer study and that uniformity along lines of latitude for normal eggs may make it possible to reduce considerably the number of readings required and thus enable far more eggs to be considered in a given time. In the light of the present work, the following problems seem to be worthy of consideration:

(1) The problem of uniformity of pattern for eggs laid by the same bird and differences between birds.

(2) The problem of the pattern as hens build up the shell and whether this is constant at all stages of shell formation.

(3) The problem of changes in thickness during incubation.

(4) The problem of the thickness pattern and cracking. In relation to this last problem, it is well known that most measurements of breaking strength have been related to mean shell thickness or to some criterion assumed to represent this, such as percentage shell or the specific gravity of the egg. The considerable variations in thickness in different parts of the same shell suggest that some parts may crack far more easily than others and hence mean thickness and general breaking strength may give quite misleading results. Work is already in progress on this problem.

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Dept. of Physiological Chemistry
The University
Reading

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STUDIES ON EGG SHELLS. XVII.*—Variations in Membrane Thickness and in True Shell Nitrogen over different Parts of the same Shell

By C. TYLER

Variations in membrane thickness and in total, insoluble and soluble nitrogen in the true shell over different parts of the same shell have been considered.

For membrane thickness the variations around any particular latitude were much smaller than variations along any longitude. Variations around any one collar were greater for membrane thickness than for shell thickness and greater for total nitrogen than for membrane thickness.

* Part XVI: preceding paper

Longitudinal patterns of membrane thickness were generally typical for any one bird and, in fact, variation from bird to bird occurred almost entirely between the waist and the narrow pole of the egg. From the broad pole to the waist, practically all eggs showed a decreasing membrane thickness. Membrane thickness in some areas of an egg can be twice as great as in other areas. In adjacent areas of shell, membrane thickness tends to be significantly correlated, but for areas some distance apart there is no such correlation.

The amount of total, insoluble and soluble nitrogen in the true shell showed no general longitudinal pattern.

There was no relationship between membrane thickness and shell thickness in different parts of the same shell, nor for total shell nitrogen and shell thickness.

Introduction

This paper deals with the variation in thickness of the shell membrane and in the total, insoluble and soluble nitrogen content of the true shell.

The method of obtaining values for the membrane has been fully described by Tyler,[1] but it should be stressed that, whereas for shell thickness each of the 50 pieces of shell gave six thickness readings, the pieces only give one reading for the membrane. It is not possible, therefore, to go into the same detail for membrane thickness as it is for shell thickness. The membrane thickness has been expressed as μg. of nitrogen/cm.^{2}, instead of converting the values to protein. Forty-six eggs laid over a short period of time and taken from six birds were available.

The total, insoluble and soluble nitrogen in the true shell have been obtained by methods previously described.[2] As the size of the individual pieces of shell is too small to permit of the determination of both total and insoluble nitrogen, four alternate pieces of each collar have been used for the one determination and the remaining four for the other, while soluble nitrogen was found by difference. Eighteen eggs from two birds were available. The shells used for the true shell nitrogen determinations had previously been used for measurements of shell thickness[3] and hence it was possible to consider the relationship, if any, between them.

Birds 500, 502, 503 laying normal eggs and bird L laying thin-shelled eggs provided both shell thickness and membrane thickness values[3] and again any relationship could, therefore, be studied.

Results

Membrane thickness (μg. of N/cm.^{2})

Analysis of variance showed that, as with shell thickness, the degree of variation around any one collar is very small compared with the considerable variations from pole to pole. Therefore, the mean of each of the eight latitudinal values has been taken and these six mean values, plus the single value for each cap, give the pattern of variation from pole to pole. Clearly the single values for the caps are less reliable than the mean values for the latitudinal collars.

Before considering these results, however, it is of interest to consider the coefficients of variation between the membrane thicknesses in any one collar. A typical set of values has been given in Table I and it will be seen that these values are much greater than the ones for shell thickness in normally shelled eggs.[3] Further, the thin-shelled eggs give larger coefficients of variation than the normally shelled eggs. Of course, this may be a matter of individual bird variation, and it will be necessary to compare the membranes of normal and thin-shelled eggs from the same bird before reliable conclusions can be drawn.

There were no differences of pattern between clutches of eggs from the same bird, therefore the eggs for each bird have merely been numbered in sequence.

The individual patterns are shown in Figs. 1–12 and it will be seen that, apart from one or two exceptions, the pattern for the eggs of any one bird is similar. Generally speaking, in the case of birds G, L and 503, each pattern shows that the broad pole has the thickest membrane, the narrow pole has a thinner membrane and somewhere between and usually near the equatorial region, the thinnest membrane of all occurs. For most eggs bird H shows a constantly decreasing thickness from the broad pole down to the collar adjacent to the narrow pole cap. Then at the pole itself there is a slight increase. Birds 500 and 502 show a pattern

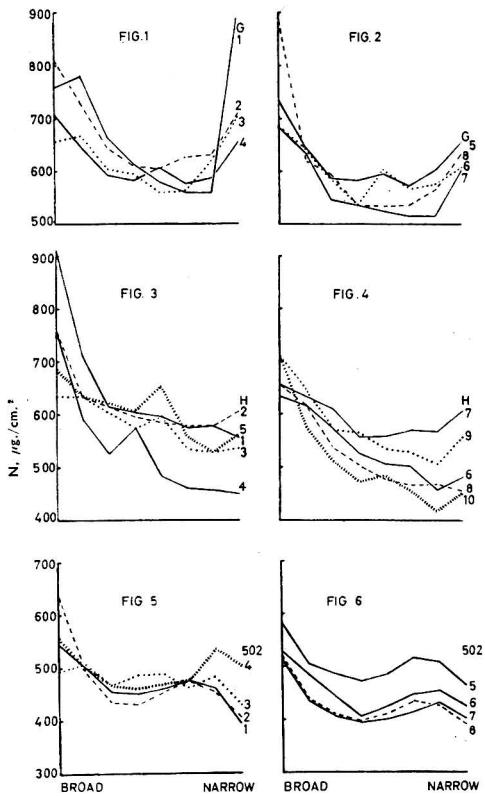
Table I

Membrane thickness (μg. of N/cm.²)

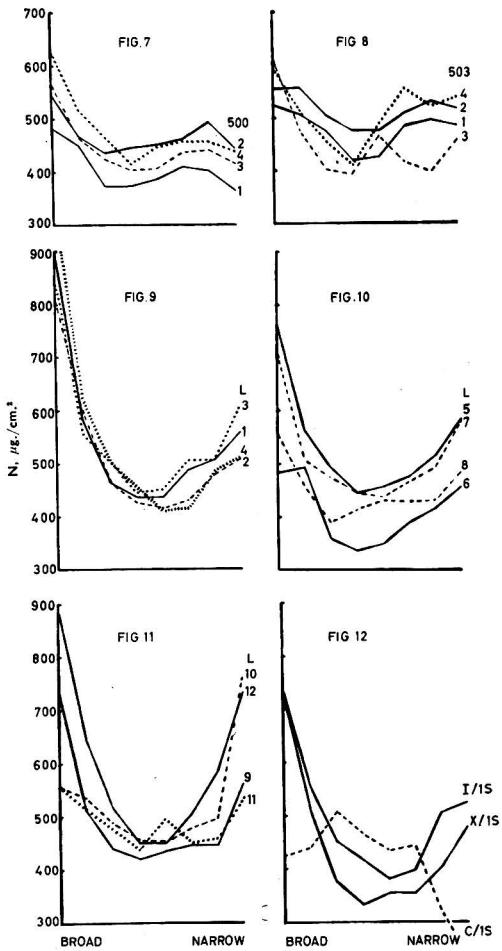
Coefficients of variation between the eight values of any one latitudinal collar
G and H gave normally shelled eggs, L and C abnormally thin-shelled eggs

Collar	Bird			
	G	H	L	C
B	2·61	2·99	6·97	5·29
C	5·66	3·09	4·86	12·01
D	2·92	2·68	2·68	6·46
E	4·65	1·37	5·99	5·99
F	2·08	4·48	5·37	23·47
G	3·18	3·07	5·98	17·66
Mean	3·52	2·95	5·64	11·81

very similar to the group G, L and 503, except that the narrow pole has a thinner membrane than the collar adjacent to it. All six birds show a similar pattern of continuing thinning of the membrane as far as the equator and, in fact, only eight eggs out of 46 show even the slightest deviation from this pattern. It is only from the equator to the narrow pole that real variations in pattern occur.



FIGS. 1–6.—*Variations in membrane thickness (μg. of N/cm.²) from the broad to the narrow pole of normally shelled eggs*



FIGS. 7–12.—*Variations in membrane thickness (μg. of N/cm.²) from the broad to the narrow pole of eggs*
Figs. 7 and 8 normally shelled eggs, Figs. 9–12 abnormally thin-shelled eggs

The eggs considered here from bird L were thin-shelled as also were the single eggs from birds X and I (Fig. 12). These eggs all showed the general pattern, but the thin-shelled egg from bird C gave a different pattern.

The other major point to notice is the very large differences which may occur in membrane thickness over different parts of the same shell. With bird L, for example, the maximum values at the broad pole are usually one and a half times to twice as great as the minimum value, somewhere near the waist, and with bird G, two eggs show differences of one and a half times.

For birds 500, 502, 503 and L, values for both shell and membrane thickness within the same egg were available. A study of these values for individual eggs showed that there was no sign of a relationship between them.

True shell nitrogen (parts per 100,000)

When the results for individual birds were subjected to an analysis of variance it was found that the variation from pole to pole was always greater than that around any collar. In some cases the variation from pole to pole was highly significant when compared with error but in other cases this was not so, while the variation within a collar was never significantly greater than error.

Further, it was not possible to trace any general pattern in the distribution of total nitrogen in the shell from pole to pole even for eggs from the same bird. These remarks also apply to insoluble and soluble nitrogen and, therefore, no results have been given.

Attempts to relate the values for soluble and insoluble nitrogen for any one egg were unsuccessful as was an effort to relate total shell nitrogen to shell thickness within one egg.

Discussion

The results show that the latitudinal variation is much less than the longitudinal one with membrane but the position with regard to true shell nitrogen is very variable. The membrane values, in general, give a picture of decreasing membrane thickness from broad pole to equator, but beyond this to the narrow pole, different birds show somewhat different patterns. It is not yet possible to discuss the reasons for this in detail until more eggs have been studied, but it may relate to the mechanism of membrane formation.

The lack of a relationship between shell and membrane thickness within eggs suggests that the membrane already formed has no influence on the formation of the shell which follows. This is, of course, not surprising since the two are formed in entirely different parts of the oviduct and are of quite different materials.

On the other hand, the meagre evidence available suggests that thin-shelled eggs have membranes which show greater latitudinal variation than those which are normally shelled, but clearly more data is required.

In the paper dealing with shell thickness values,[3] it was shown that there was a definite relationship between thickness in different parts of the shell and that those caps and collars nearest to each other showed the highest degree of correlation with the exception of cap H. The twelve eggs from bird L were used for a similar calculation relating to membrane thickness (see Table II). It will be seen that adjacent caps and collars are significantly correlated, but that for areas distant from each other, there is generally no significant correlation. Only collar C is significantly correlated with all others. A selection of correlation coefficients was also calculated for the eight eggs of bird G and for the ten eggs of bird H. The values (not given) support the general picture found for the eggs of bird L. Thus it would appear that the very close relationships found for shell thickness do not hold for membrane thickness. Again this may be a result of entirely different structures, mode of formation, or some other factor, but it is not possible to say until more work has been done on membrane and shell formation.

Total, insoluble and soluble nitrogen show no real general pattern from one pole of the egg

Table II

Correlation coefficients for the relationships between the thickness of membrane in different caps and collars

Bird L: 12 eggs

	A	B	C	D	E	F	G	H
A	—	0·840***	0·647*	0·560	0·075	0·338	0·607*	0·116
B	0·840***	—	0·739**	0·519	0·110	0·389	0·779**	0·378
C	0·647*	0·739**	—	0·902***	0·648*	0·709**	0·813***	0·644*
D	0·560	0·519	0·902***	—	0·761**	0·683*	0·677*	0·584*
E	0·075	0·110	0·648*	0·761**	—	0·686*	0·419	0·489
F	0·338	0·389	0·709**	0·683*	0·686*	—	0·803**	0·770**
G	0·607*	0·779**	0·813**	0·677*	0·419	0·803**	—	0·750**
H	0·116	0·378	0·644*	0·584*	0·489	0·770**	0·750**	—
Mean	0·445	0·536	0·729	0·669	0·455	0·625	0·693	0·533

*** Significant at $P < 0.001$ ** Significant at $P < 0.01$ * Significant at $P < 0.05$

The remaining values are not significant

to the other and even for individual eggs the pattern is of little consequence, because latitudinal variations are often nearly as great. Further, there is no relationship between true shell nitrogen and shell thickness; it would thus appear that the finding of Tyler & Geake[4] that there is an inverse relationship between shell thickness and true shell nitrogen when large numbers of eggs are considered, does not hold within any one egg.

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Dept. of Physiological Chemistry
The University
Reading

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CHEMICAL STUDIES ON THE HERRING (*CLUPEA HARENGUS*). V.*—The Effect of Heat Processing on the Extractable Nitrogen Fraction

By R. B. HUGHES

The non-coagulable nitrogen fraction of herring increases during heat processing, being influenced by processing time and temperature. Free amino-acids did not contribute to this increase, which was shown to be due to (*a*) gelatin derived from connective tissue, (*b*) ammonia derived from unknown sources, (*c*) nitrogenous components which became available for extraction during heat processing and which yielded ammonia under conditions of acid hydrolysis. The significance of these results in relation to quality in canned herring is discussed.

Introduction

When herring flesh is macerated with dilute aqueous trichloroacetic acid solution, proteins are coagulated and low-molecular-weight nitrogenous components such as free amino-acids, lower peptides, trimethylamine oxide, creatine, ammonia, etc., are extracted. The total nitrogen in such an extract is termed the total extractable nitrogen (T.E.N.) fraction of the fish. The object of the work presented in this paper was to investigate the extent and nature of the changes which occur in this fraction when herring are heat processed.

There is little published work available on this subject. Mudrak[1] studied the behaviour of carp flesh when cooked in brine and in brine containing acetic acid, and Bendall[2] examined the changes occurring in lean beef during heat processing at various temperatures.

Experimental

Both fresh and frozen herring were used in these studies. The fresh fish were either used within 24 h. of capture or were frozen in an air-blast freezer, stored at −29°, and allowed to thaw

* Part IV: *J. Sci. Fd Agric.*, 1960, **11**, 700

at room temperature before use. Herring were heat-processed in various ways. Duplicate blocks of flesh from single fish were cut as described previously,[3] one being cooked in sealed glass tubes in an oven, and the other analysed raw for purposes of comparison. The cooked blocks rested on small glass stools so that the exuded liquor could be drained off separately. Nobbed (beheaded and eviscerated) herring were packed in lacquered 14-oz. oval fish cans, the cans exhausted by heating for 10 min. in a steam chest at 98°, seamed and processed in an autoclave. When it was necessary to isolate the exuded liquor, a piece of wire gauze partitioned off a small section at one end of the can. After being processed, the can was punctured at this end and the liquor drained off. The lid was then removed and the cooked fish withdrawn in the normal manner. Minced herring flesh was cooked in 14-oz. oval fish cans.

Preparation of extracts

Unless otherwise stated, extracts were prepared with a 10% solution of trichloroacetic acid in water (hereafter referred to as 10% T.C.A.). The material to be extracted was mixed thoroughly and a 20-g. sample extracted with three successive 50-ml. portions of 10% T.C.A. in an M.S.E. Nelco homogeniser. The extracts were centrifuged and the separated liquid layers bulked and made up to 200 ml. with 10% T.C.A. The solution was filtered and either analysed immediately or stored at −29° pending analysis.

Estimation of total nitrogen in the extracts

Borate buffer solution.—Boric acid (10 g.) was dissolved in 700 ml. of distilled water and 200 ml. of ethanol and 10 ml. of mixed indicator solution were added. The colour was adjusted to the desired faint reddish-brown with dilute sodium hydroxide solution before finally making up to 1 l. with distilled water.

Mixed indicator solution.—Toshiro's indicator was prepared by dissolving 0·033 g. of bromocresol green and 0·066 g. of methyl red in 100 ml. of ethanol.

A 5-ml. aliquot of the T.C.A. extract was digested for 5 h. in a micro-Kjeldahl flask with 5 ml. of conc. sulphuric acid, 0·5 g. of selenium dioxide and a tablet containing 1·25 g. of potassium sulphate and 0·15 g. of mercuric sulphate. The digest was made up to 50 ml. with distilled water and a suitable aliquot transferred to a Markham micro-distillation unit, made alkaline with 10 ml. of 40% sodium hydroxide solution containing 4% of sodium thiosulphate, and steam distilled for 7 min. into 10 ml. of borate buffer. Back-titration was carried out with standard 0·01N-hydrochloric acid, delivered from a micro-burette. Duplicate estimations were carried out on all solutions analysed, and 100% recovery was obtained when amino-acid nitrogen was estimated by this method on a solution having a total nitrogen content similar to that of the flesh extracts.

Estimation of the free amino-acids, taurine and ammonia

Separations were carried out on 150-cm. and 15-cm. columns of Amberlite I.R.G.-120 resin at 50° as described by Moore *et al.*[4] Two-ml. samples of eluate were collected and analysed as described in the photometric ninhydrin method of Moore & Stein,[5] except that 0·08% of stannous chloride was added as reducing agent to the ninhydrin reagent instead of hydrindantin. Also the ninhydrin solution was de-mineralised before use by treatment with ion-exchange resin as described by Jacobs.[6] In practice it was found that 10% T.C.A. extracts could not be applied directly to the 150-cm. columns in the required amount (10 ml.) because the separation of glycine and alanine was affected, due presumably to the trichloroacetic acid disturbing the pH condition on the columm. Partial neutralisation of the extract with sodium hydroxide solution overcame this difficulty, but caused the volume to become too large for convenient application to the column. Extracts prepared with 5% instead of 10% T.C.A. gave good separations of glycine and alanine, and as there was no difference between the nitrogen content of extracts prepared with the two concentrations of T.C.A., a 5% solution was used for preparing extracts required for column chromatography.

Results quoted for methionine include the sulphoxide, and no correction is made for the slight loss of serine and threonine which is reported to occur on the column.

In exploratory runs on synthetic amino-acid mixtures in 5% T.C.A., satisfactory recoveries (±5%) of amino-acids were obtained. Proline and hydroxyproline were not estimated by this method, but independently by the methods described below.

Hydroxyproline was estimated by the method of Neuman & Logan.[7] It was found necessary to extend the initial heating and shaking time to 20 min. to ensure complete removal of excess peroxide. The estimation was not affected by the presence of T.C.A. The red colour formed was measured in a photoelectric colorimeter with Kodak No. 5 filters (max. transmission at 590 mμ). Recoveries of known amounts of hydroxyproline added to T.C.A. extracts were not less than 90% at low concentrations, and normally higher than 95%.

Proline was determined by Troll & Lindsley's method.[8] A weakly acidic ion-exchange resin (Zeokarb 226, H^+ form) was used to remove interfering basic amino-acids from the solutions instead of Permutit as suggested by the authors. This treatment caused no loss of proline when carried out in neutral solution, but a loss occurred when the medium was acidic, as was the case with T.C.A. extract. It was therefore necessary to remove the T.C.A. before carrying out the analysis. This was done as follows: a 10-ml. aliquot of the extract was passed down a short column (1·5 cm. × 1 cm.) of Amberlite I.R.G.-120 (200 mesh, H+ form), and the column washed free of T.C.A. with water. The amino-acids were displaced with 20 ml. of 2N-aq. ammonia and the ammonia removed by boiling. This solution was then treated with Zeokarb 226 without loss of proline. In the case of hydrolysates, where the T.C.A. decomposed and hydrochloric acid was removed by evaporation, it was not necessary to treat with Amberlite I.R.G.-120. Colour intensity was measured in a photoelectric colorimeter and Kodak No. 4 filters (maximum transmission at 540 mμ). 100% recovery of proline was obtained on known additions made to flesh extracts.

Hydrolysis of T.C.A. extracts

An aliquot of the solution, normally 2 ml., was placed in a Pyrex test tube with an equal volume of conc. hydrochloric acid, and the tube sealed and heated in an oven at 116° for 24 h. The tube was then opened* and the contents transferred to a small conical flask and evaporated to dryness in a vacuum desiccator over solid sodium hydroxide. The residue was dissolved in water and aliquots taken for analysis as required.

Preparation of hydrolysed samples of herring skin and scales

Skin was removed from the back and sides of 57 herring, the fish having just been scrubbed with a nail brush to remove the scales as completely as possible. The skins were washed with water, acetone (three times) and ether (three times). Eight g. of dry skin were obtained in this way. Scales were also treated in a similar manner.

Skin (or scales) (0·5 g.) was hydrolysed by heating at 116° in a sealed glass tube with 20 ml. of 6N-hydrochloric acid. The resulting solution was filtered, made up to a suitable volume with distilled water and aliquots taken to dryness in a vacuum desiccator over solid sodium hydroxide to remove hydrochloric acid. The residue was dissolved in the minimum volume of water and applied to the chromatography column.

Results and discussion

Blocks of herring flesh were cooked in sealed glass tubes and the cooked flesh and exuded liquor analysed separately. The corresponding uncooked block in each case was also analysed for comparative purposes. The results obtained are shown in Fig. 1, and all are expressed on a raw-weight basis. It will be seen that the T.E.N. content of the cooked flesh dropped initially compared with that in the raw flesh, but this drop did not continue, the T.E.N. content of the cooked flesh remaining about 100 mg./100 g. lower than that of the raw flesh even after being cooked for 5 h. T.E.N. in the exuded liquor rose steadily over the period, and this resulted in an overall increase in T.E.N. (cooked flesh and liquor), compared with the uncooked flesh. Similar results were obtained when nobbed unbrined herring were canned (4 fish per 14 oz. oval can) and processed at 116° for periods of up to 5 h. Three cans were processed for each period of

* Care must be taken when opening these tubes because of the high pressure formed by decomposition of T.C.A. to chloroform and carbon dioxide

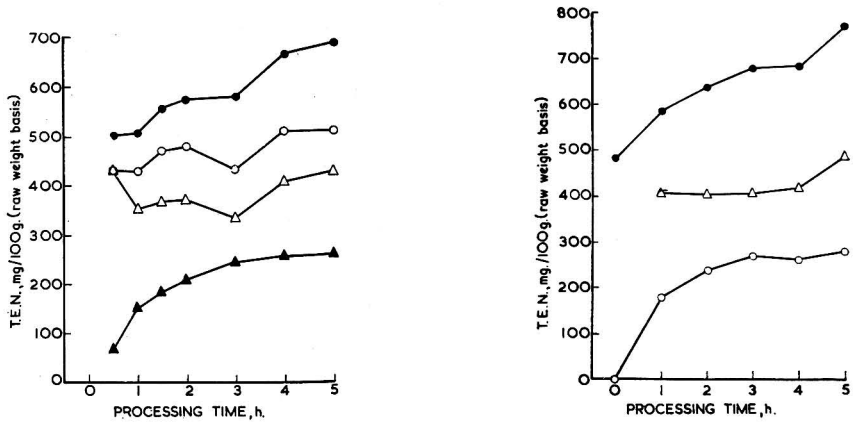

FIG. 1.—*Production of extractable nitrogen (T.E.N.) in herring blocks processed in sealed tubes at* 116°
● total (cooked + liquor) ○ raw block △ cooked block ▲ exuded liquor

FIG. 2.—*Production of extractable nitrogen (T.E.N.) in unbrined nobbed herring processed in oval cans at* 116°
● total △ cooked fish ○ liquor
(each point represents the mean value for three cans analysed. Mean deviation not greater than ±3% of mean value)

time, and the contents of each analysed, six raw nobbed fish were minced, well mixed and three samples extracted and analysed. Fig. 2 shows that although the drained flesh did not increase markedly over the 5 h. period, the increase in the T.E.N. content of the exuded liquor again caused an overall steady increase in T.E.N. This increase continued even after processing for 11 h. as shown in Fig. 3, where the T.E.N. content of the cooked flesh (including liquor) is given.

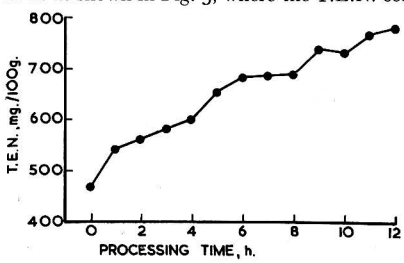

FIG. 3.—*Production of extractable nitrogen (T.E.N.) in unbrined nobbed herring processed in oval cans at* 116°
(each point represents the analysis of one can)

These experiments therefore established that an increase in T.E.N. occurs when herring are heat-processed, and that this increase is found in the exuded liquor. A similar increase was reported by Bendall[2] when lean beef is pressure cooked at 126° for 3 h. ; most of the components causing the increase in extractable nitrogen could be dialysed. Bendall therefore suggested that the increase might be due to the degradation of proteins to lower peptides or free amino-acids. It was therefore decided to investigate whether part or all of the increase in T.E.N. which occurs in herring could be accounted for by the production of free amino-acids.

Minced herring flesh was processed in 14-oz. oval cans at 116° for periods up to 3 h., and extracts prepared in 5% T.C.A. The amino-acid analyses obtained on these extracts are shown in Table I,* which also includes the figures for ammonia. Fifteen amino-acids (including taurine) were present in the raw fish. Arginine was also present in a concentration of approximately 20 mg./100 g., but was not estimated in this series of experiments. Later work (see Table II)* showed that its concentration was not affected by heat processing. An account of some of the free amino-acids present in herring flesh has been given previously.[9] The present results provide a more complete picture of the free amino-acid pattern. It will be seen that no increase occurred in any of the amino-acids during cooking that would account for the increase

*The results in Tables I–III were obtained in one series of experiments in each case, to reveal trends as the processing times increased

in T.E.N. The only amino-acid which showed a regular change as cooking progressed was histidine, which decreased over the period. This was also observed when herring blocks were cooked in sealed glass tubes and the extract prepared with 80% ethanol. The reason for this loss is not yet known. Histidine is reasonably heat-stable, and the loss must be due either to its interaction with other flesh components, or to adsorption effects similar to these observed in the case of creatine.[10] The increase in ammonia which occurs during cooking has been reported previously.[3] This would only account for a fraction of the increase in T.E.N.

Table I

Effect of heat processing at 116° on free amino-acid content of herring

Amino-acid	Amino-acid, mg. per 100 g. of nobbed fish				Amino-acid	Amino-acid, mg. per 100 g. of nobbed fish			
	Period of cooking, h. at 116°					Period of cooking, h. at 116°			
	0	1	2	3		0	1	2	3
Taurine	179	174	187	178	Methionine	7·6	8·7	9·6	9·2
Threonine	20·5	19·8	19·8	21·5	Isoleucine	12·5	9·1	14·7	13·9
Serine	17·3	17·2	18·7	21·4	Leucine	25·6	25·1	24·9	24·1
Glutamic acid	27·1	26·1	23·7	25·3	Tyrosine	14·2	12·3	13·4	13·6
Proline	19·9	16·3	16·9	17·6	Phenylalanine	13·5	11·2	12·8	12·5
Glycine	25·1	23·9	21·8	24·3	Lysine	47·6	56·8	68·8	62·6
Alanine	51·2	57·6	58·9	53·7	Histidine	108·0	59·1	43·4	23·1
Valine	19·8	21·7	24·8	30·4	Ammonia	14·4	37·2	38·4	47·1

As free amino-acid production was not the cause of the increase in T.E.N., further experiments were carried out to discover whether lower peptides were formed during cooking. Unbrined nobbed herring (which had been cold-stored and thawed before use) were processed in oval cans (five fish per can) at 116° for 1-, 3- and 5-h. periods, and extracted with 10% T.C.A. Duplicate extracts were hydrolysed and analysed for amino-acids and ammonia. The unhydrolysed extracts from the raw sample and the sample cooked for 5 h. were also analysed before hydrolysis. Mean results are shown in Table II and on a nitrogen basis in Table III.

Comparison of the unhydrolysed extracts of the raw (0 h.) fish and the 5-h. fish showed that cooking had no significant effect on the free amino-acid content (with the exception of histidine), thus confirming the results of Table I. Comparison of the hydrolysed extracts from the four samples shows that apart from histidine (which dropped steadily) and taurine (which remained unchanged) a steady increase occurred in the case of the other acids, thus suggesting that one or more peptides was being produced in increasing amount as cooking proceeded. The increase was greatest in the case of glycine, alanine, proline, glutamic acid, aspartic acid and arginine, while hydroxyproline, absent in the extract from raw fish, also increased steadily. The appearance of this amino-acid, together with large increases in the other acids mentioned above, suggested that they were being produced from the hydrolysis of gelatin or other peptides derived from the further degradation of gelatin itself. The amino-acids mentioned above are major components of fish collagens and gelatin[11] while hydroxyproline, so far as is known, is a component only of connective tissue proteins. Thus it would appear that part at least of the increase in T.E.N. which occurs during cooking is due to the conversion of connective tissue collagen to gelatin which is soluble in aqueous trichloroacetic acid. The gelatin may be further degraded to lower-molecular weight components during extended cooking at these high temperatures. As previously mentioned, it was found[2] that the increase in T.E.N. from beef was dialysable, and as gelatin was not dialysable, it was concluded that the increase would not be due to this. Gelatin degrades further during cooking, however, and it is possible that the products of gelatin cooked at 126° for 3 h. would be largely dialysable.

Reference to Table III shows that the increase in amino-acid nitrogen between 0 h. (hydrolysed) and 5 h. (hydrolysed) was 115·4 mg./100 g., while the increase in ammonia nitrogen accounted for 76 mg., giving a total increase of 191·4 mg./100 g. The increase in T.E.N. as measured directly by digestion and distillation was 208 mg./100 g., so that 92% of this increase is accounted for by gelatin nitrogen and ammonia nitrogen. Referring again to Table II, it will be seen that the ammonia content of the unhydrolysed extract from the uncooked herring

Table II

Free amino-acids and ammonia (mg./100 g. nobbed fish) in unhydrolysed and in hydrolysed extracts of herring processed at 116°

Period of cooking, h.	Taurine	Aspartic acid	Threonine	Serine	Glutamic acid	Proline	Hydroxy-proline	Glycine	Alanine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Phenyl-alanine	Lysine	Histidine	Arginine	Ammonia	Total
0[a]	191·5	3·9	8·4	9·2	13·4	N.D.	nil	18·4	34·7	10·9	4·7	5·9	8·5	4·9	6·9	29·2	156·0	14·2	19·6	—
0[b]	203·7	10·2	11·4	11·1	26·8	16·4	nil	114·5	38·0	10·9	nil	6·3	11·1	7·2	14·4	33·1	148·c	13·3	81·5	181·3
1[b]	188·0	38·4	19·9	21·3	53·1	56·2	19·2	166·9	73·3	24·1	nil	7·3	17·6	4·4	12·1	45·7	117·5	32·5	126·0	248·1
3[b]	220·0	71·6	36·3	36·7	98·9	101·7	38·8	220·0	98·3	32·4	nil	20·1	36·6	8·1	31·4	61·9	88·7	54·0	121·8	291·1
5[b]	206·0	97·5	48·2	55·2	128·0	128·3	43·6	277·0	128·6	46·0	nil	33·2	52·9	15·5	31·2	75·9	71·9	78·9	170·8	372·7
5[a]	203·0	N.D.	12·5	11·7	17·3	N.D.	nil	21·7	39·0	13·6	8·5	7·5	11·6	11·4	8·9	27·7	79·4	9·8	61·0	—

Table III

Free amino-acid and ammonia nitrogen (mg./100 g. nobbed fish) in unhydrolysed and hydrolysed extracts of herring processed at 116°

Period of cooking, h.	Taurine	Aspartic acid	Threonine	Serine	Glutamic acid	Proline	Hydroxy-proline	Glycine	Alanine	Valine	Methionine	Isoleucine	Leucine	Tyrosine	Phenyl-alanine	Lysine	Histidine	Arginine	Ammonia	Total
0[a]	21·45	0·41	0·99	1·23	1·28	N.D.	nil	3·42	5·46	1·30	0·44	0·63	0·91	0·38	0·59	5·59	42·25	4·56	16·1	—
0[b]	22·80	1·07	1·34	1·48	2·55	2·00	nil	21·35	5·98	1·30	nil	0·67	1·19	0·58	1·22	6·34	40·10	4·28	67·0	181·3
1[b]	21·08	4·40	2·34	2·84	5·06	6·84	2·05	30·04	11·52	2·88	nil	0·78	1·88	0·34	1·02	8·76	31·82	10·44	104·0	248·1
3[b]	24·62	7·52	4·27	4·89	9·42	12·35	4·14	41·05	14·46	3·88	nil	2·14	3·91	0·63	2·66	11·87	24·00	17·70	100·6	291·1
5[b]	23·08	10·26	5·66	7·36	12·20	15·62	4·66	51·65	20·21	5·50	nil	3·55	5·66	1·20	2·64	14·55	19·48	25·38	143·0	372·7
5[a]	22·72	N.D.	1·47	1·56	1·64	N.D.	nil	4·05	6·14	1·63	0·80	0·80	1·24	0·88	0·75	5·32	21·51	3·15	50·2	—

a unhydrolysed *b* hydrolysed N.D. not-determined

was 19·6 mg./100 g. while that from the fish cooked for 5 h. was 61·0 mg./100 g. The increase represents ammonia produced from unknown sources during cooking. Hydrolysis of the uncooked extract caused an increase in ammonia of 81·5 − 19·6 = 61·9 mg./100 g. which represents ammonia formed by hydrolysis (or decomposition during hydrolysis) of extractable components present in the cooked fish. Hydrolysis of the 5-h. extract caused a much greater rise in ammonia; 170·8 − 61·0 = 109·8 mg./100 g. The difference between the increase on hydrolysis of the 5-h. extract compared with the uncooked extract (109·8 − 61·9 = 47·9 mg./100 g.) represents 'hydrolysis-produced' ammonia derived from components which became available for extraction during cooking. It is unlikely that the hydrolysis of gelatin would produce this amount of ammonia. Table V shows the results obtained when whole herring skin and scales were analysed. It will be seen that an abnormally large quantity of ammonia was produced when the skin was hydrolysed, suggesting that there are components in the skin which yield ammonia under these conditions. It is possible that these components become increasingly available for extraction along with the gelatin during cooking, and this could account for the 'hydrolysis-produced' ammonia which is formed during cooking.

Table IV

Comparison of rates of production of T.E.N. and gelatin (as hydroxyproline) in herring processed under various conditions

(results as mg./100 g.)

Condition of fish	Processing temp., °F	Processing time, h.	Increase in T.E.N. (A)	Increase in hydroxyproline (B)	Ratio A/B
Nobbed	230	1	7*	1·6*	4·4
Skinned	240	1	19	4·1	4·6
after	250	1	36	7·9	4·5
brining	260	1	56	12·3	4·6
Nobbed	220	1	8·5	2·0	4·3
Brined	230	1	26·5	6·2	4·3
	240	1	58·5	14·0	4·2
	250	1	87·5	23·3	3·8
	260	1	131·0	33·5	3·9
Nobbed	240	0·5	37†	9·7†	3·8
Unbrined	240	1·5	84	28·5	3·9
	240	2·5	131	34·3	4·0
	240	3·5	160	40·9	3·9
	240	4·5	184	48·2	3·9

* Increase compared with figure at 212° F
† Increase compared with 0 h. processing

Table V

Amino-acids and ammonia produced by hydrolysis of herring skin and scales

(results g./100 g. dry weight)

Amino-acid	Skin	Scales	Amino-acid	Skin	Scales
Aspartic acid	4·58	4·07	Hydroxyproline	3·56	4·47
Threonine	1·65	2·07	Proline	7·04	8·49
Serine	2·65	3·61	Tyrosine	0·78	1·85
Glutamic acid	5·53	5·95	Phenylalanine	2·24	2·50
*Glycine and alanine	16·43	16·99	Methionine	1·27	1·51
Valine	2·08	1·86	Lysine	2·79	3·18
Isoleucine	1·27	0·97	Histidine	0·89	1·21
Leucine	1·45	2·30	Arginine	4·08	3·44
			Ammonia	3·44	0·75

* As glycine

An interesting point which emerges from Table I can be seen by comparing the glycine content of the hydrolysed and unhydrolysed extracts from the uncooked fish. A large increase occurs which suggests that there is a peptide present in the fish with glycine as its major component.

Further evidence that the increase in T.E.N. which occurs during cooking is derived from changes in the connective tissue is given by the results of the following experiments.

Thawed frozen-stored July herring were nobbed and brined (20 min. in saturated salt solution) and one lot of fish was then skinned. The fish were packed in oval cans and processed at temperatures varying from 100° to 125°. Six cans were processed at each temperature. The content of each can was well mixed and a sample from each extracted. The extracts from pairs of cans were bulked, the three bulked extracts being analysed. In addition to determination of T.E.N. the extracts were hydrolysed and hydroxyproline estimated. As this amino-acid is derived from connective tissue proteins, it can be used as an indirect measure of the gelatin present in an extract. The results obtained are shown in Fig. 4. Increasing the processing temperature causes a marked increase in the rate of production of both T.E.N. and gelatin, while the effect of removing the skin on the production of T.E.N. and of gelatin is apparent. Fig. 5

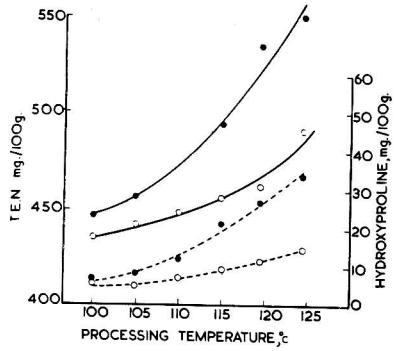

FIG. 4.—*Production of extractable nitrogen (T.E.N.) and gelatin in brined nobbed herring processed in oval cans for 1 h.*

T.E.N. with skin
„ without „
hydroxyproline with skin
„ without „

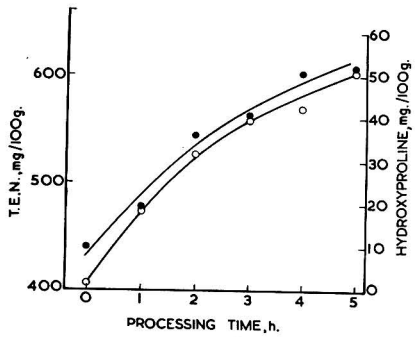

FIG. 5.—*Production of extractable nitrogen (T.E.N.) and gelatin in unbrined nobbed herring processed in oval cans at 116°*

● T.E.N. ○ hydroxyproline

(Each point represents the mean value of six cans analysed. Mean deviation for hydroxyproline is less than the diameter of the points, and for T.E.N. not greater than ±2% of the mean value)

shows the production of T.E.N. and gelatin when herring are processed over a prolonged period at one temperature. Again six cans were processed for each period of time, and extracted and bulked as described above. Six raw fish were minced, well mixed and three samples extracted and analysed. It is obvious from these results that there is a close relationship between the increase in T.E.N. occurring during cooking, and the degradation of connective tissue collagen, and this is confirmed by the figures (derived from Figs. 4 and 5) given in Table IV, which show that the ratio (increase in T.E.N./increase in hydroxyproline) remained quite constant irrespective of the time or temperature of processing.

Mudrak,[1] studying the behaviour of carp flesh cooked in brine, found that the proportion of non-protein nitrogen to total (water) soluble nitrogen increased only slightly when the processing temperature was increased from 100° to 117°, but showed a much greater increase when the temperature was increased to 126°. This was due largely to an increase in the ' intermediate products of protein degradation ' (i.e., non-coagulable nitrogen less amino-nitrogen and volatile basic nitrogen). He suggested therefore that at temperatures above 117°, hydrolysis of flesh proteins becomes more extensive and unspecified drastic undesirable changes begin to occur in the flesh. It is evident from Fig. 4 that the production of non-coagulable nitrogen in herring also increases rapidly as the processing temperature is raised. It is not likely, however, that the nature of the changes which occur varies with increase in temperature. If this were so, production of non-coagulable (extractable) nitrogen from sources other than collagen would show up at higher processing temperatures by causing an increase in the T.E.N. hydroxyproline ratio.

The results of the experiments described in this paper suggest that the increase in T.E.N.

which occurs when herring are heat-processed is caused by (*a*) gelatin which is derived mainly from the skin, (*b*) ammonia which is produced from unknown sources during heat processing and (*c*) unidentified nitrogenous components which yield ammonia under conditions of acid hydrolysis and which may be associated with the connective tissue proteins of the skin. It is not known whether the conversion of collagen to gelatin is important with regard to flavour. Gelatin itself is comparatively tasteless, but under the processing conditions used for herring, considerable degradation of the gelatin to smaller peptide fragments may occur. This aspect requires further study. There is no doubt, however, that the production of gelatin is important with regard to the texture of the fish. The herring is held together structurally by the connective tissue of the skin, muscle and bone. The conversion of this to gelatin with consequent loss of holding ability results in the cooked fish being softer and more liable to break up on handling. Therefore measurement of hydroxyproline production (itself an index of the degree of conversion of collagen to gelatin) may serve as an objective method for measuring the tendency for the fish to break up after heat processing. The possibility is being investigated of using this method in studies of factors which may effect the firmness and appearance of canned herring.

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Torry Research Station
Aberdeen

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TREATMENT OF MEATS WITH IONISING RADIATIONS. VII.*—EFFECT OF LOW TEMPERATURES DURING IRRADIATION

By B. COLEBY,† M. INGRAM, H. J. SHEPHERD, M. J. THORNLEY and G. M. WILSON‡

Raw pork and beef have been irradiated with 2 MeV electrons at controlled temperatures from +18° to −196°. Appearance, odour and flavour were assessed by a taste panel, the destruction of glutathione was estimated chemically; and the extent of the survival of the bacteria determined. In each case, a relation to the temperature of irradiation was observed similar to that previously recorded for thiamine. There was little effect between 18° and 0°; but a rapidly increasing protection from 0° to −20°, with a smaller increase down to −196°. Experiments in the range 0° to −10°, in which destruction of glutathione was the criterion, indicated that protection began when ice separated from the tissue.

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† Present address: Office of the Scientific Adviser, North Atlantic Treaty Organisation, Paris XVI, France

‡ Present address: Mars Ltd., Slough, Bucks.

It is suggested, accordingly, that the degree of protection depends on the amount of free water withdrawn from the system. The importance of precisely defining temperature during irradiation is emphasised. It is concluded that freezing will be of little benefit in radiation pasteurisation processes aimed at destruction of vegetative micro-organisms; but that it might be helpful in radiation sterilising processes, as the protection is smaller for spores.

Introduction

Adverse quality changes of considerable magnitude are produced in foods by irradiation, and methods to prevent them have received much attention. Early publications[1, 2] indicated that freezing during irradiation was very effective but this was not confirmed in later reports.[3–7] More recent investigations[8–11] in this laboratory have repeatedly shown that meats can be considerably protected by freezing during irradiation. Comparisons made with beef and pork, given various doses of radiation at room temperature or at −75°, have revealed a marked protection in samples irradiated at −75°, both in respect of quality and of chemical changes: the degree of protection varied according to the criterion studied, but was usually found to be greater than about 2·5, i.e., a sample of meat given 5 Mrads at −75° was preferred to one given 2 Mrads at room temperature. It has also been found, however, that the actual temperature during irradiation (i.e., the degree of freezing) influences the degree of protection from chemical change;[10, 11] and in the present paper, further results are presented which show the efficacy of various temperatures below freezing in affording general protection to beef and pork during irradiation. Changes in quality, chemical composition and survival of micro-organisms have been studied.

Experimental

Materials

Fillets of pork and rump steak were purchased from a local retailer a few days after the animals had been slaughtered. The meat was prepared and packed anaerobically in shallow aluminium cans (1·2 cm. in depth) as described previously.[8] Cans were kept at 0° overnight before being placed at the desired temperature (at least 2 h. before irradiation).

Irradiation

Temperatures were controlled by immersing samples in eutectic mixtures or, occasionally, freezing mixtures.[12] During irradiation, the cans were supported so that the upper surfaces just emerged from the surface of the coolant. Samples were irradiated with the scanned electron beam from a Van de Graaff generator (2 MeV). The required dose was absorbed in increments of not more than 1 Mrad spaced at intervals of about a minute, and cans were irradiated from opposing sides successively. When accurate control of temperature was desired, the radiation was spaced out over longer intervals.

Quality assessment was made by a taste panel, as previously described.[13] The panel normally had ten members who were discriminating, but not all especially familiar with irradiation flavours.

Chemical estimations

Glutathione was estimated by a method similar to that of Batzer & Doty.[14] Minced meat (10 g.) was weighed accurately into a beaker, and about half of it transferred to a vortex flask. Metaphosphoric acid (2·25% 35 ml.) was added, the mixture homogenised for 1 min., and sodium chloride (15 g.) was then added to the mixture, which was homogenised for a further minute. The contents were decanted into a volumetric flask, and the process repeated with the remainder of the minced meat. These operations were performed at 5°. The volume of the mixture was adjusted to 100 ml. with 2·25% metaphosphoric acid, and the mixture was well shaken and left for 10 min. After filtration (Whatman No. 50 paper), 8 ml. of the filtrate were allowed to react with 4% sodium nitroprusside solution (1 ml.) and then with 1·5M-sodium carbonate/0·033M-sodium cyanide solution (1 ml.). The absorption was measured as quickly as possible in an EEL colorimeter, with filter Ilford 624 (500–540 mμ).

pH was measured after homogenisation of 8 g. of meat in 16 ml. of M/200-sodium iodoacetate for 2 min. at room temperature. A glass electrode was used.

Microbiological examinations

Only beef was used for these experiments and it was prepared as described above, then frozen at −20°, and brought to the required irradiation temperature at least 2 h. before the experiment. Control samples were treated similarly to ensure that any effects of the freezing itself were common to all samples.

After irradiation, 5-g. samples were removed from each can and homogenised with 45 ml. of quarter-strength Ringers solution.[15] Serial ten-fold dilutions were made from this suspension with 0·1% peptone water.[16] Portions of 0·33 ml. of appropriate dilutions were transferred to heart infusion agar (Difco) plates, previously surface-dried, and spread over one quarter of the surface. Where small numbers of survivors were expected, 0·5-ml. portions of the initial suspension were spread over whole plates. The plates were incubated for 2 days at 20°.

Results

Organoleptic changes

In raw minced pork.—The results of panel assessments of changes in appearance, odour and flavour of raw pork irradiated at various temperatures are shown in Table I. It is apparent that the quality changes were markedly less in those samples irradiated while frozen. Generally, the quality was better at lower temperatures, although the protection increased most rapidly at temperatures just below freezing point down to about −20°. Additional protection was sometimes conferred by irradiating samples at −196°, rather than −80°, but the results as a whole suggest that the benefit of this additional cooling is comparatively small.

Table I

Panel assessment of (a) *raw minced pork samples,* (b) *raw minced beef samples irradiated at various temperatures*

R = average rank S = average hedonic score

Temp. during irradiation	Appearance R	Odour R	Flavour R	Flavour S
1 Mrad	(a)			
0°	4·6	4·85	4·0	5·35
−5°	2·4	3·7	3·55	5·6
−10°	2·45	3·1	2·95	5·9
−20°	2·1	2·15	2·1	6·35
−75°	3·45	1·2	2·4	6·45
No. on panel	13	13	20	
Significance†	***	***	***	
4·5 Mrads	(b)			
Control††	3·0	1·0	1·0	7·2
18°	4·5	5·0	4·9	2·6
−20°	2·75	4·0	3·7	4·0
−75°	2·0	2·5	2·6	5·0
−196°	2·75	2·5	2·8	5·3
No. on panel	4	4	10	
Significance	n.s.	***	***	
3 Mrads	(a)			
Control††	1·0	1·0	1·3	6·3
18°	4·7	4·8	4·6	3·0
−20°	4·0	4·2	4·2	3·5
−75°	3·3	3·0	2·4	5·5
−196°	2·0	2·0	2·5	5·5
No. on panel	6	6	10	
Significance	***	***	***	
2 Mrads	(b)			
0°	5·0	4·8	4·1	4·25
−5°	2·6	3·4	3·1	4·9
−10°	2·7	3·1	3·3	4·8
−20°	2·8	2·1	2·2	5·65
−75°	1·9	1·5	2·3	5·55
No. on panel	21	21	40	
Significance	***	***	***	

† The statistical significance of ranking data in this and other Tables was evaluated as described by Kendall[31]

†† Controls unirradiated and stored at −20°

n.s. = not significant * = significant at 5% level

** = significant at 1% level *** = significant at 0·1% level

In raw minced beef.—Similar comparisons made with samples of raw beef irradiated at various temperatures are also recorded in Table I. Again, the degree of protection was related to the temperature of the sample during irradiation, in the same general way as for pork.

In precooked minced beef and pork.—Table II presents data on panel assessment of the quality changes in precooked pork and beef samples irradiated at low temperatures. As an additional protective measure, ascorbic acid (0·1% by weight) was incorporated as a solution into some of the samples of minced meat shortly before freezing. This combination of treatments largely offset the quality changes induced by irradiation: indeed, on occasion, samples irradiated at −196° were ranked as favourably as control samples.

Table II

Panel assessments of (a) *pre-cooked minced pork samples,* (b) *pre-cooked minced beef samples irradiated at various temperatures, with and without the addition of ascorbic acid (AA)*

R = average rank S = average hedonic score

Temp. during irradiation	Appearance R	Odour R	Flavour R	Flavour S	Temp. during irradiation	Appearance R	Odour R	Flavour R	Flavour S
3 Mrads	(a)				*3 Mrads*	(a)			
Control†	1·3	1·7	1·1	7·0	Control†	2·2	1·2	2·3	5·45
18°	4·7	5·0	4·7	3·4	18°	3·8	4·0	4·0	2·8
−20°	4·3	3·8	3·8	4·5	−75° + AA (0·1%)	2·4	3·0	2·3	4·5
−75°	3·0	3·2	2·8	5·5	−196° + AA (0·1%)	1·6	1·8	1·4	5·9
−196°	1·7	1·3	2·6	6·1	No. on panel	5	5	10	
No. on panel	6	6	10		Significance	*	***	***	
Significance	***	***	***						
4·5 Mrads	(a)				*4·5 Mrads*	(a)			
Control†	1·0	1·0	1·4	6·3	Control†	1·6	1·0	1·5	5·8
18°	5·0	5·0	4·7	2·8	18°	3·6	4·0	3·8	2·6
−20°	2·25	3·5	3·6	4·2	−75° + AA (0·1%)	3·2	2·8	2·5	4·75
−75°	3·25	3·0	3·0	4·7	−196° + AA (0·1%)	1·6	2·2	2·2	5·2
−196°	3·5	2·5	2·3	5·3	No. on panel	5	5	10	
No. on panel	4	4	10		Significance	**	***	***	
Significance	***	***	***						
3 Mrads	(b)				*4·5 Mrads*	(b)			
Control†	1·8	1·0	1·6	6·35	Control†	1·3	1·0	1·7	5·5
18°	3·5	4·0	4·0	3·5	18°	3·8	4·0	4·0	2·4
−75° + AA (0·1%)	2·3	3·0	2·4	5·0	−75° + AA (0·1%)	2·7	2·8	2·5	4·4
−196° + AA (0·1%)	2·3	2·0	2·0	5·7	−196° + AA (0·1%)	2·2	2·2	1·8	5·5
No. on panel	6	6	10		No. on panel	6	6	10	
Significance	n.s.	***	***		Significance	***	***	***	

† Controls unirradiated and stored at −20°

Chemical changes

Changes in pH.—The changes of pH in minced beef and pork after irradiation with 5 Mrads at various temperatures are shown in Table III. The changes brought about by irradiation were small, but freezing diminished them. With beef, the lower the temperature during irradiation the smaller was the change; with pork no obvious trend appeared.

Table III

Changes in the pH (Δ pH) of raw minced pork and beef resulting from the absorption of 5 Mrads at various temperatures

Temp. during irradiation	Pork		Beef	
	Initial pH	Δ pH	Initial pH	Δ pH
18°	6·30	+0·19	5·58	+0·22
0°	6·30	+0·11	5·55	+0·19
−3°	6·31	+0·01	5·56	+0·18
−12°	6·31	+0·09	5·55	+0·17
−25°	6·32	+0·08	5·55	+0·15
−38°	6·32	+0·07	5·57	+0·13
−58°	6·32	+0·10	5·56	+0·13
−75°	6·32	+0·10	5·55	+0·13

Destruction of glutathione.—The percentages of glutathione remaining in samples of minced raw beef and pork after doses of 5 Mrads at various temperatures are shown in Table IV. Control experiments confirmed that the freezing did not itself measurably affect the glutathione, but freezing before irradiation markedly protected it, particularly if very low temperatures were used. The similarity in the degree of protection afforded in both beef and pork is noteworthy. The destruction of glutathione at temperatures just below freezing is recorded in Table V. For both beef and pork, protection did not become apparent until the temperature

was between −3·2° and −6·0°. Of the water in meat, about 75% is frozen at −3° and 98% at −20°, hence it seems likely that freezing of water is probably responsible for a large part of the protection observed. Other experiments[17] have shown that, with beef and pork, the point at which protection becomes apparent lies between −3° and −6° if the meat is slowly cooled to this temperature; on the other hand, if the meat is first frozen, say at −20°, and then allowed to warm up to almost 0°, protection is retained until about −1°, again substantiating the belief that the freezing of water in the tissue is an important factor.

Table IV

Retention of glutathione in raw minced beef and pork irradiated at various temperatures

(dose 2·5 Mrads)

Temp. during irradiation	Glutathione retained, %	
	Pork	Beef
20°	40	41
0°	42	43
−3°	57	53
−12°	75	57
−25°	78	74
−38°	81	81
−58°	81	77
−75°	84	83
−196°	95	98

Table V

Retention of glutathione in raw minced beef and pork by irradiation at temperatures near 0°

(dose 2 Mrads)

Temp. during irradiation	Glutathione retained, %	
	Pork	Beef
18°	35·9	36·8
0°	36·8	37·9
−1·1°	35·9	32·1
−2·6°	37·5	30·5
−3·2°	35·9	30·5
−6·0°	56·6	40·7
−10·6°	59·5	50·0

Inactivation of micro-organisms

Fig. 1 shows the effect, on the numbers of surviving micro-organisms, of irradiation with a constant dose of 0·2 Mrad at various temperatures. The control points were provided by samples taken through the same cycle of temperature changes without being irradiated. They show a wide scatter, as expected for a mixed population of organisms in minced meat, but no indication of any significant damaging effect due to the various temperature changes themselves. Counts on the irradiated samples indicated a protection by irradiating at temperatures below 0°, increasing considerably down to about −20°. The proportion of survivors at −20° was about the same as at −80°. There was no difference between the number of survivors after irradiation at 0° and +18°, and this was confirmed in another experiment.

The types of micro-organisms present were not precisely determined, though they were mostly bacteria. Judging from other experiences (e.g., [18]), they would presumably have been largely in the vegetative state, and their behaviour was consistent with that view.

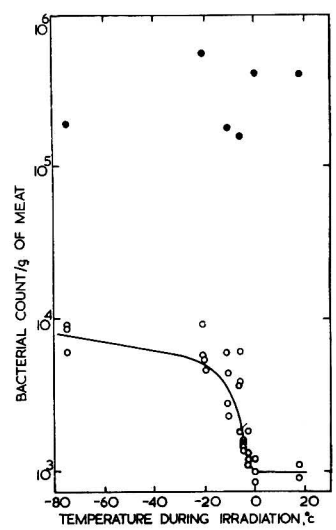

FIG. 1.—*Effect of temperature during irradiation with a dose of 0·2 Mrad, on survival of the normally-occurring bacteria, in minced beef packed under N_2*

(individual points represent replicate experiments)
● control ○ irradiated with 200,000 rads

Discussion

Considered as a whole, the results presented here amply confirm the earlier observations. With each character investigated, there appeared the same general pattern as was evident in

our first experiments on the influence of irradiation temperature:[8, 10, 11] little effect of temperature in the 'normal' range of 0° to + 20°, with a rapidly increasing protection by lowering the irradiation temperature from freezing point to about −20°, and only slightly greater influence at still lower temperatures. This is the pattern with characters as different as the glutathione content and the number of surviving bacteria.

Further, even though panel assessments cannot be considered as a sensitive analytical tool, the same pattern of differences in quality can be distinguished between samples irradiated at various temperatures below freezing. On balance, it appears that irradiation of samples while frozen in liquid nitrogen, results in the least quality change, but a large proportion of the protection is already achieved with temperatures as high as −20°. Even at this latter temperature, the gain may be by a factor of several-fold; and in this way, on favourable occasions, we have for the first time been able to apply near-sterilising doses to raw meats, without making them (in our view) unacceptable. The idea of irradiating food at temperatures of about −20° does not seem hopelessly impractical with refrigeration facilities currently available. Irradiation of samples while frozen does not of course eliminate all changes in quality. Obviously, too, the procedure is not universally applicable; for example with fruits, freezing would of itself cause extensive damage; while with other foods, such as frankfurters[6] and sausages,[19] freezing before irradiation does not seem to confer any protection, for reasons still unknown.

Because the degree of protection varies so much with the degree of freezing, it is obviously important in experimental work to define the temperature during irradiation. Failure to do this may explain why others have not always observed gains in quality by irradiating below freezing point: for instance, in the experiments of Schultz *et al.*,[6] the temperature of samples during irradiation was certainly above −20°, and it seems probable that it might have been nearly 0°.

The destruction of glutathione is a convenient chemical measure of damage which occurs during irradiation, and such changes may be observed more precisely than may quality changes using a taste panel. Others have shown that there is a good correlation between chemical changes and quality changes, both immediately after irradiation[20] and during storage;[21] and it is clear, from comparison of the behaviour of glutathione and thiamine[10, 11] with that of the organoleptic changes in our experiments, that the same correlation holds in this effect of freezing.

Using destruction of glutathione as an index of quality change, a more detailed investigation was made of the exact temperature at which protection becomes effective (Table V). This supports the belief that an important part of the protection is due to freezing of the free water in the tissue; for by cooling beef and pork to the desired temperatures, protection was never apparent until the temperature was less than −3°, while if the meat was first cooled to a lower temperature, protection was still observed with samples irradiated at −1°. It is reasonable to suppose that this hysteresis phenomenon results from supercooling of water and retention of crystalline structure, respectively. The destruction of glutathione by irradiation at various temperatures resembles closely the behaviour commonly observed in the irradiation of dilute aqueous solutions in frozen conditions. The protection afforded by freezing seems to become effective quite rapidly, since Hannan & Shepherd[22] observed no differences in behaviour between samples which had been maintained at temperatures between −10° and −70° for periods from 1 h. to 6 days before irradiation at these temperatures.

Although crystallisation of water may be responsible for some of the protection, freezing will also alter the rates of reaction of radicals formed by irradiation, and hence the general course of subsequent reactions. The importance of defining the precise temperature during irradiation has been demonstrated with such a simple system as hydrogen peroxide solutions,[23] where a change of temperature from −196° to −180° during irradiation considerably affected the reaction. Similarly, Hannan[24] found that irradiated fats gave widely varying yields of peroxides, depending on the temperature during irradiation and subsequent storage: this was attributed to variations in the reactivity of radicals at different temperatures, a supposition later confirmed by O'Meara & Shaw by use of electron spin resonance techniques.[25] With a more complex system containing yeast cells and cysteine, Smaller & Avery[26] also noticed differences in the rates of decomposition of radicals as the temperature of irradiated samples was raised towards the

freezing point. It is reasonable to suppose that similar effects occur with foodstuffs, so that although a given dose of radiation may produce similar numbers of ionised and excited atoms in frozen and unfrozen samples, the radicals subsequently formed may differ in quantity and quality in the two cases. Presumably some radicals react much more readily at low temperatures than do others, and hence the course of reactions occurring as the sample is thawed may differ completely from those occurring in samples irradiated while unfrozen. The striking differences in quality and chemical composition of samples irradiated at different low temperatures amply testify to the profound differences in reaction pattern.

Regarded in this way, freezing during irradiation is equivalent to disturbing the time sequence of reactions during irradiation, which suggests that large alterations in dose rate might produce similar effects, with correspondingly beneficial effects. It is worth recalling that the original publications of Brasch & Huber[1, 27] did, of course, stress the importance of using extremely high dose rates (10^{12} rads per sec.) to prevent excessive quality changes on irradiation.

The data in Fig. 1 confirm that the naturally-occurring micro-organisms in meat will also be protected by freezing, in the same manner and to about the same degree as the organoleptic characters. Further, although our data are inadequate to show them, the observations of Wood[28] on vegetative yeast cells demonstrated the same supercooling phenomena as were recorded above with glutathione. Because the protection of vegetative micro-organisms by freezing is thus essentially the same as the protection of quality in meat, it appears that the increased dose required microbiologically might about cancel any gain in quality due to irradiating while frozen. In this context, however, the type of micro-organisms to be inactivated must be considered. Vegetative bacteria are usually protected by freezing;[29] therefore, with pasteurising treatments (<1 Mrad) where vegetative bacteria are the main target, the gain in quality due to irradiating while frozen may well be offset by the higher dose needed to inactivate the bacteria, as was indeed indicated in earlier experiments with low dose treatments of minced chicken meat.[15] On the other hand, bacterial spores, although variable in their response to temperature of irradiation,[30] are not generally much protected by freezing, so that for sterilisation procedures, where the aim is to kill spores, the gain due to freezing should be considerable, as we have already suggested elsewhere.[9]

Acknowledgment

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Low Temperature Research Station
Downing St.
Cambridge

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DIFFUSION OF GLUCOSE DURING VEGETABLE DEHYDRATION

By R. B. DUCKWORTH and G. M. SMITH

Strips of potato and carrot were soaked in a solution containing glucose labelled with ^{14}C until the distribution of the labelled glucose was uniform through the material. The strips were then dehydrated, with and without a prior scald, and changes in the distribution of glucose studied by autoradiography. In scalded potato and carrot strips there is a net inward movement and the resulting accumulation of glucose in the centre is considered to explain the greater susceptibility of this region to non-enzymic browning. In unscalded potato strips, a peripheral concentration of glucose develops.

Introduction

It has recently been shown by the use of autoradiography[1] that sulphite, applied to strips of root vegetables in scalding solutions, diffuses through the whole volume of the strip during subsequent dehydration. The greater susceptibility of the central part of the strip to browning during drying and storage at elevated temperatures, is not due to an inadequate penetration of sulphite, since, in the dried strip, the sulphite accumulates in this central region. These results appeared to lend some support to an earlier contention[2] that naturally occurring solutes, such as sugars and amino-acids, would themselves migrate towards the centre, due to the formation of a concentration gradient resulting from the more rapid removal of water from the surface layers. A central accumulation of such substances and perhaps also of colourless intermediate compounds formed during the progress of the browning reactions, might indeed result in a more rapid browning of this central region. It was decided therefore to study the diffusion of glucose in such materials under similar conditions.

Experimental

Radioactive materials

Glucose, randomly labelled with carbon-14, was obtained from the Radiochemical Centre, Amersham. This was used to prepare a solution containing 0·31 mmoles of glucose per ml. and having an activity of 0·5 μc/ml. Strips of potato and carrot $\frac{3}{16}$ in. $\times$ $\frac{5}{16}$ in. in cross section were soaked in this solution for 40 h., after which time the distribution of the labelled glucose was virtually uniform through the material.

Processing

Strips of potato and carrot prepared as above were scalded for 3 min. in boiling water before dehydration. In addition, strips of potato were dehydrated without a prior scald. Drying was carried out on a wire mesh tray in an electrically-heated oven, the temperature of which was maintained at 100° for 40 min., 75° for a further 2½ h. and 65° for a final period of 2½ h.

Samples of six strips were taken immediately after scalding and after drying respectively for 40, 60, 80, 120, 180, 240, 300 and 340 min. On removal from the oven each strip was quickly weighed, then immediately frozen in acetone–solid CO_2 and finally freeze-dried. The freeze-dried strips were transferred to a vacuum desiccator and stored over P_2O_5 until they had reached constant weight. This enabled the moisture content of each strip at the time of sampling to be calculated.

Autoradiography

In order to obtain flat sections suitable for autoradiography it was necessary to embed the material in a suitable medium. Strips which had been dried for 80 min. or less were sufficiently soft to be embedded in paraffin wax for sectioning. Later samples were more brittle and required embedding in a cold-setting resin. Transverse sections 0·5–2 mm. thick were cut from the embedded strips and placed in contact with a fast *X*-ray film. The films were developed after exposure for 7 days at −4° in a light-proof container.

Results

Similar results were obtained with scalded material of both potato and carrot. After scalding (Fig. 1*a*), the autoradiographs show no evidence of depletion of glucose from the surface layers due to leaching and the distribution of glucose remains generally uniform. During the first 80 min. of drying, the distribution of glucose did not depart from the uniform in any

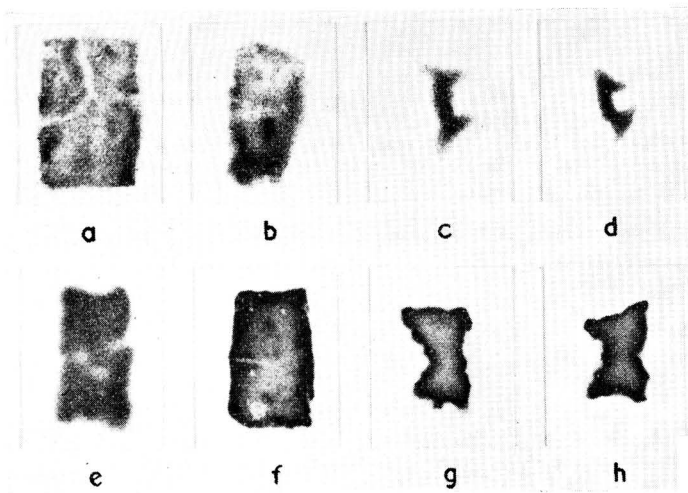

FIG. 1.—*Autoradiographs of transverse sections of potato strips taken at different stages during the dehydration process and showing the distribution of* ^{14}C*-labelled glucose*

	Time of drying	Moisture content
(*a*) Scalded potato	immediately after scalding	80·4%
(*b*) Scalded potato	1 h.	58·7%
(*c*) Scalded potato	2 h.	41·8%
(*d*) Scalded potato	5 h.	7·7%
(*e*) Unscalded potato	before drying	81·5%
(*f*) Unscalded potato	1 h.	64·7%
(*g*) Unscalded potato	2 h.	47·1%
(*h*) Unscalded potato	5 h.	9·2%

characteristic way, though individual sections show local pockets of higher activity (Fig. 1*b*). After drying for 2 h. (Fig. 1*c*), a central accumulation of glucose is now evident and the marked shrinkage of the strip is illustrated by the much smaller area of the autoradiographic image. The pattern of distribution remains similar throughout the remainder of the drying cycle (Fig. 1*d*).

Changes in the distribution of glucose within unscalded potato strips during drying are in marked contrast to those taking place in the scalded material. After drying for only 40 min., a higher concentration of glucose could be detected at the periphery of the strip and this peripheral concentration became intensified during the first 2 h. of drying (Fig. 1*f*, g). No further change in distribution took place during the later stages of drying (Fig. 1*h*).

Discussion

The contrast shown between the behaviour of glucose in scalded and unscalded potato is probably due to the difference in form of the starch in the two cases. During the early stages of drying in unscalded material, in which the starch grains remain intact, films of water containing the glucose are present in capillary spaces between the closely packed grains. As water is removed rapidly from the surfaces of the strips, these films will be drawn outwards carrying the glucose by mass flow. Outward movement of water in the gelatinised starch of the scalded material will be initially much slower.

The present results confirm that the predominant direction of diffusion of solutes during the dehydration of scalded strips of potato and carrot is towards the centre of the piece. There seems little doubt that the resulting accumulation of reactive substances is responsible for the greater susceptibility of this part of the strip to non-enzymic browning. The redistribution of glucose, like that of sulphite,[1] is brought about quite early during the drying cycle. The observed pattern of distribution does not change after the first 2 h. of drying, although it is known from other experiments, the results of which will be reported later, that glucose can diffuse slowly through such materials at moisture contents much lower than those normally reached at this stage. There must, however, be a critical level of moisture below which diffusion of solutes cannot take place. If the final moisture content is lower than this critical value, the irregular distribution of solutes observed at the end of drying will be maintained. It seems unlikely in such a case that reactions between sugar and amino-acid molecules could be initiated during storage. Browning, under these conditions, is presumably limited to the formation of colour in self-browning intermediates formed at an earlier stage. If the final moisture content is now lower than the critical value, a further slow redistribution of solutes will occur during subsequent storage and browning reactions will proceed more readily.

Dept. of Food Science
The Royal College of Science and Technology
1 Horselethill Road
Glasgow, W.2

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JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE

ABSTRACTS

JUNE, 1961

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.

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ERRATUM

J.S.F.A. Abstracts, 1961, i.

Col. 210, line 5 for *J. Dairy Sci.* read *Agricultura.*

JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE

ABSTRACTS

JUNE, 1961

I.—AGRICULTURE AND HORTICULTURE

General: Soils and Fertilisers

Soils of South Tunisia. H. N. le Houérou (*Ann. agron.*, 1960, **11A,** 241—308).—A survey of geological features and classification of the soils with special reference to their water-retaining capacity. (59 references.) P. S. Arup.

Pedogenic and petrogenic characteristics of soil profiles developed in silt-mantled acid shale. E. Wurman (*Soil Sci.*, 1960, **90,** 348—356).—Two soil profiles developed under silt-mantled acid shale are examined. The soil formation is discussed in detail and it is apparent that the underlying material has had a major part in determining the characteristics of the solum. T. G. Morris.

Statistical parameters and reproducibility of the neutron method of measuring soil moisture.—J. F. Stone, R. H. Shaw and D. Kirkham (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 435—438).—Using the neutron method for determining soil moisture, coeff. of variation for reproducibility (repeat measurements at a given location) was about 1·5% for both laboratory and field readings. The neutron method required about 14% of the sampling sites of the gravimetric method to give equal standard errors of the mean for each method. A. H. Cornfield.

Slow tests under soil moisture suction. J. L. McMurdie and P. R. Day (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 441—444).—Standard Slow Tests (standard strength test of saturated cohesive soil using the triaxial compression apparatus) on two soils and glass beads were compared with Slow Tests wherein part of the stresses were applied in the form of soil moisture suction. The differences obtained indicated that soil moisture suction is not equivalent, in general, to mechanical stresses as regards the strength property. A. H. Cornfield.

Hygrophotographic measurement of soil moisture following protracted drought. J. Sivadjian (*Soil Sci.*, 1960, **90,** 369—373).—Hygrophotographic plates (*ibid.*, 1957, **83,** 109—112) were used during a prolonged drought in 1959 to measure the moisture content of garden soil in Paris. The method of calibrating the plates is described in detail. Results show that after a 4-month drought, the moisture at 12 in. depth is enough to release 0·18 μg. per sq. mm. per min. of water to the gelatin of the plate. T. G. Morris.

Piezometric field study of soil water movement towards tile drains in a Nappanee silty clay loam. Clyde L. Wilson (*Dissert. Abstr.*, 1960, **21,** 713—715).—Descriptions are given of normal flow-patterns and their modifications by sloping ground, variations in the height of the water table, and lateral seepage from undrained ground. P. S. Arup.

Drainable porosity evaluation from outflow measurements and its use in drawdown equations. G. S. Taylor (*Soil Sci.*, 1960, **90,** 338—343).—A theoretical and mathematical discussion. T. G. Morris.

Plant and moisture relationships with vegetable crops on a light sandy soil. D. W. Stolp (*Versl. landbouwk. Onderz.*, 1960, 66.16, 235 pp.).—The literature of the subject is critically reviewed. The responses observed during three seasons of a large no. of market-garden crops to different regulated levels of average soil moisture are compared; differences are examined with respect to crop characteristics, the root system, climatic conditions and time of harvesting. In general, an average of ~12·5% of soil moisture is required for max. yields on this type of soil; this corresponds with a max. drying-out of 11·5% of the available moisture (the difference between field-capacity and the wilting level), or a moisture retention of pF = 2·5. Critical periods of drought-susceptibility arise through root-failure at times of intense top-growth. The relative importance of average soil moisture, water retention (pF) and the extent of depletion of the available moisture is considered. (258 references.) P. S. Arup.

Changes in the pore-space of a pasture topsoil under animal treading. M. W. Gradwell (*N.Z. J. agric. Res.*, 1960, **3,** 663—674).—The content of pores in the top 1½ in. of silt loam drained at a suction of 50 cm. of water, was determined and also the vol. of water drawn from saturated cores by this suction. Least drainage was found after heavy grazing by sheep in wet weather; in one case the content of pores fell from 10 to 3% of the soil vol. but rose again following drier weather. Soil cores containing the greatest wt. of roots had the highest measured vol. of pores; they also showed a tendency to drain more than cores with fewer roots. (14 references.) E. M. J.

A vibro-compaction method for greenhouse soil-structure studies. N. J. Rosenberg (*Soil Sci.*, 1960, **90,** 365—368).—The method utilises a vibrating (3450 v.p.m.) table to which is clamped a pot containing the soil container. This is rigidly fastened to the pot and can be covered with a lid. Compaction of soil using the vibrating method gives a uniform mass with no air holes or layers of soil of different density. By varying the time of vibration and the moisture content of sandy loam it was possible to obtain plugs of soil with from 34 to 51% of total pore space and a high degree of uniformity. T. G. Morris.

Ploughpan investigations at the Great Plains field stations. L. F. Locke, H. V. Eck, B. A. Stewart and H. J. Haas (*U.S. Dep. Agric. agric. Res. Serv.*, 1960, Prod. Res. Rep. 40, 33 pp.).—Observations and data pertaining to root impedance, infiltration penetration, bulk density, soil moisture, org. matter, tillage and plant growth studies in the greenhouse are reported for the stations at Woodward, Okla., and Mandan, N. Dak. (10 references.) E. G. Brickell.

Use of sodium tripolyphosphate as dispersing agent in the mechanical analysis of soils. E. Jouis, M. T. Lecacheux and F. Cauchy (*Ann. agron.*, 1960, **11,** 231—240).—The polyphosphate ($Na_5P_3O_{10}$) solution (20 g./l.) is prepared at room temp., filtering if necessary; it is stable for several weeks. The soil sample (10 g. of clay or 20 g. of sandy soil) is shaken mechanically with 100 ml. of the dispersant. Soils containing <3% of $CaCO_3$ and >2·5% of org. matter are subjected to preliminary treatment with H_2O_2. A correction is made for the wt. of dissolved polyphosphate present in the separated fractions of soil. The procedure is suitable for distinguishing between calcareous and siliceous sands. A. G. Pollard.

An oscillating permeameter [for soils]. E. C. Childs and A. Poulovassilis (*Soil Sci.*, 1960, **90,** 326—328).—The measurement of the hydraulic conductivity of soil is briefly discussed and a method using an oscillating apparatus described. T. G. Morris.

Mutable factors affecting yields on loess-loam soil.—J. Livens (*Agricultura*, 1960, **8,** 177—202).—A review covering measures for maintaining the structure and humus content of the soils, and factors influencing the development of the root system. (22 references.) P. S. Arup.

Factors affecting the removal of salts from halloysite. G. W. Thomas (*Soil Sci.*, 1960, **90,** 344—347).—The silt and clay fractions of two hydrated halloysites from two soils were saturated with either KCl or NaCl and then washed with water, methanol or ethanol and the amount of salt remaining estimated by Cl^- analysis. Na-saturated halloysite was almost free of salt after two washings with methanol. K-saturated halloysite previously oven dried was washed free almost as easily, but the unheated mineral was much more difficult to wash free of salt with methanol, while halloysite, KCl-saturated and then oven dried, was the most difficult. The replacement of KCl from hydrated halloysite was rapid with water, slower with methanol and very slow with ethanol. The results are discussed in relation to the high cation-exchange capacities usually reported for hydrated halloysite. T. G. Morris.

Hydrolysis of aluminium salts in clay and soil systems. J. L. Ragland and N. T. Coleman (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 457—460).—Al salts underwent more extensive hydrolysis in acid soil and clay systems than in aq. solutions of the same pH, presumably due to sorption of hydrolysis products on the soils and clays. During the "increased hydrolysis" reaction 3 moles of H^+ appeared for each mole of Al hydrolysed and sorbed. When $AlCl_3$-treated soils or clays were treated with dil. HCl, 3 moles of H^+ were required to dissolve or render exchangeable each mole of Al. This indicated that the sorbed material was $Al(OH)_3$, although calculations of solubility products indicated that it was not gibbsite. A. H. Cornfield.

Measurement of exchangeable aluminium in soils and clays. C. Lin and N. T. Coleman (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 444—446).—Neutral N-NaCl, -KCl and -$CaCl_2$ leached Al^{3+} from Al-saturated soils and clays in amounts approx. equiv. to the exchange capacity of the Al-saturated materials. N-KCl extracted about the same amounts of Al^{3+} from soils and clays with extractant pH ranging from 2 to 7. N-$CaCl_2$ extracted $Al^{3+} + Ca^{2+} + Mg^{2+}$ (mequiv.) from acid subsoils in amounts equiv. to the exchange

capacity of the soils. Potentiometric titrations of Al-saturated or naturally acid soils and clays to pH 6 in N-KCl consumed quantities of base equiv. to the Al displaced by salt leaching or determined by conductimetric titrations. With soils containing mostly kaolin minerals and Fe oxides the acidity which could be titrated was larger than the amount of Al which could be displaced.
A. H. CORNFIELD.

Some effects of irrigation waters of differing quality on soil properties. R. L. Hausenbuiller, M. A. Haque and A. Wahhab (*Soil Sci.*, 1960, **90,** 357—364).—The correlation between the "residual Na_2CO_3" in irrigation water and the level of exchangeable Na in the soils to which it was applied was highly significant. High correlations were also found between the exchangeable Na in the soil and the % of sol. Na in the water. Correlations between conductivities of the irrigation water and the saturation paste extract were not significant. Grass was grown in columns of soil in cement pipes and irrigated with a simulated saline water. With water having an Na/Ca ratio of 1 : 1, exchangeable Na accumulated to a larger extent in the subsoil than in the surface. With a 9 : 1 ratio exchangeable Na tended to accumulate in the upper layers. Removal of Ca by water containing HCO_3^- occurred even when the sol. Na content of the water was only 50%. The length of time of contact of water and soil was of importance. T. G. MORRIS.

Adsorption and release of strontium from clays and soils with equilibration, isotopic tracer and plant uptake techniques. E. O. McLean, T. G. Arscott and V. V. Volk (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 453—457).—When added to clays ^{90}Sr was adsorbed to a smaller extent by illite than by bentonite. Uptake of Sr by soya-beans from the treated clays was greater from illite than from bentonite. Increasing the Ca- or Ba-saturation of the clays decreased Sr adsorption by the clays and increased Sr uptake by the plants. Increasing the pH of a soil from 4·7 to 7·2 by liming had little effect on the amounts of Sr adsorbed from $SrCl_2$ solution.
A. H. CORNFIELD.

Changes with time in the availability of strontium-90 in soil. H. M. Squire (*Nature, Lond.*, 1960, **188,** 518—519).—Preliminary work suggests that the availability of ^{90}Sr in soil is reduced by a small amount over a period of 3½ years; it would appear to be converted into less accessible forms. The effect of such physico-chemical changes on the absorption of ^{90}Sr by plants is too small to be of great significance in the assessment of the passage of ^{90}Sr through food chains. E. G. BRICKELL.

Diffusion of ions in soils. R. K. Schofield and I. J. Graham-Bryce (*Nature, Lond.*, 1960, **188,** 1048—1049).—Theory of a simplified method of determining the diffusion coeff. (D) of an ion through soil is given and experimental procedure is described. Two disks (1-in. dia.) of Permaplex ion-exchange membrane are equilibrated separately in identical solutions containing equal aliquots of a soil, one solution containing a radio-isotope of the specific ion. The disks (soil layer ~2·5 mm.) are removed, held in contact for ~24 h. and one disk is used to separate the active and inactive layers in a small closed cylinder (1-in. dia.), the other disk being used to check that no activity is gained or lost from the cylinder. After a known time the two layers of soil are finally extracted to determine the amount of radio-isotope diffusing from one layer to the other, and D is calculated by theory. The method obviates interference by other adsorbed cations; D can attain a max. of $1{\cdot}2 \times 10^{-7}$ cm.²/sec.
W. J. BAKER.

Secondary alkalisation processes in some irrigated areas to the East of the River Tisza. K. Darab (*Acta agron. Acad. Sci. hung.*, 1959, **9,** 363—405).—An account is given of the effects of irrigation on the mobilisation of the sol. alkaline constituents of the soils of these areas and of counter-measures which have been adopted. Rotation with ley courses has proved partly successful; in some cases it has been necessary to lower the water table by drainage and to apply chemical improvers to the soil. (57 references.) P. S. ARUP.

Influence of nature of clays on dynamics of potassium in some experimental fields. J. J. Franc de Ferrière, R. Blanchet, G. Millot and T. Camez (*Ann. agron.*, 1960, **11A,** 163—175).—The requirements of potatoes grown on chloritic clay soils, poor in K and of low retentive capacity for K, are best served by moderate applications of K at frequent intervals; these soils can be improved by the addition of humus which increases their capacity for the temporary retention of K. Heavier applications are necessary on illitic or montmorillonitic clay soils, which have a high retentive capacity for K.
P. S. ARUP.

Estimation of level of potassium reserves of soils; use of an improved Morgan–Barbier test. R. Blanchet and S. Périgaud (*Ann. agron.*, 1960, **11A,** 347—355).—The replacement of NaOAc by an equiv. amount of NH_4OAc in the extracting solution improves the accuracy of the test (especially as regards the flame-photometric determination of K) and increases the amount of K extracted from clay soils. The results of the modified test as an indication of the equilibrium between the K-reserves and the K-fixing properties of the soil, agree satisfactorily with results for the K-status of soils given by pot experiments with rye-grass and by field tests.
P. S. ARUP.

Microscopical determinations of apatite and a study of phosphorus in Nebraska soil profiles. R. F. Shipp and R. P. Matelski (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 450—452).—The technique is based on microscopical examination of the heavy soil minerals placed on 10·7N-H_2SO_4. Needle-like crystals of $CaSO_4$ develop on the surface of apatite grains. Other Ca and Mg minerals did not produce needle-like crystals. The low birefringence of apatite was a further identifying characteristic. The apatite content of a fine sand, a very fine sandy and a silty clay loam increased with depth and was correlated with acid-sol. P values. A. H. CORNFIELD.

Two methods for measuring a labile fraction of inorganic phosphorus in soils. A. van Diest, H. W. Jespersen, R. F. White and C. A. Black (*Proc. Soil. Sci. Soc. Amer.*, 1960, **24,** 498—502).—The uptake of P by sorghum from 70 soils in pot tests was predicted more precisely by P extracted from the soils with the anion-exchange resin method than the 0·03N-NH_4F–0·025N-HCl method. Modifying the former method to take into account the $H_2PO_4^-$ and HPO_4^{2-} components improved the prediction. A. H. CORNFIELD.

Effect of associated salts on transformations of monocalcium phosphate monohydrate at the site of application. D. R. Bouldin, J. R. Lehr and E. C. Sample (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 464—468).—The effect of mixing $Ca(H_2PO_4)_2,H_2O$ with various salts and placing the "granule" in a hole in a fine sandy loam (pH 5·2) on movement of P from the granule was studied. After 3 weeks at 20° the fraction of added P that remained in the granule ranged from 2% with $(NH_4)_2SO_4$ to 92% with $CaCO_3$ as the associated salt. Other salts resulted in retention of 6—27% of the added P. Uptake of P by sorghum from the soil after removal of the granule indicated that plant response was influenced by both the quantity of P transported into the soil and the composition of the solution in which the P was transported. When a calcareous silty clay loam was used the P residues in the granule were somewhat greater than with the fine sandy loam. During dissolution from the granule the soil has relatively little influence on the reactions taking place in the granule. This indicates that laboratory phase-rule studies may be useful in estimating the fraction of P remaining in the granule and the composition of the solution leaving the granule. A. H. CORNFIELD.

Forms of newly-fixed phosphorus in three acid sandy soils. T. L. Yuan, W. K. Robertson and J. R. Neller (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 447—450).—Over 80% of the PO_4^{3-} added to three sandy soils (pH 5·2—5·7) was fixed as Al and Fe phosphates and 10% was found as water-sol. and Ca phosphates. The ratio Al-fixed PO_4^{3-}/Fe-fixed PO_4^{3-} increased with rate of application of PO_4^{3-}; the extent of this effect varied with soil type. Increasing soil drying temp. (room to 100°) after treatment increased Al-fixed and decreased Fe-fixed PO_4^{3-}. Increasing the no. of cycles of wetting and drying reduced the % of Al-fixed PO_4^{3-} in two soils and increased the % of Fe-fixed PO_4^{3-} in two soils. A. H. CORNFIELD.

Influence of temperature and moisture on soil phosphorus. II. Effect prior to and during cropping on soil phosphorus availability to millet. A. R. Mack and S. A. Barber (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 482—484).—Dry matter yields and P uptake by millet increased with temp. of incubation (−20·5° to 23° for 74 days) of the soil prior to cropping. Varying the moisture content (40% to 100% field capacity) of the soil during pre-cropping incubation had little effect on yields or P uptake. When millet was grown at various soil moisture and temp. levels, P uptake increased with moisture level at the higher soil temp. (27° ± 6°), but not at the lower soil temp. (16° ± 1°). Root growth was greater at the higher soil temp., but only where P was applied. A. H. CORNFIELD.

Adsorption and isotopic dilution of phosphate ions in contact with metallic oxides and clays. R. Blanchet (*Ann. agron.*, 1960, **11A,** 55—74).—Suspensions of (undried) $Fe(OH)_3$ at pH 7·5 and especially those of $Al(OH)_3$ adsorb considerably more PO_4^{3-} from aq. Na_2HPO_4 than do suspensions of calcareous clays. In all three cases, the isotopic dilution of the adsorbed PO_4^{3-} proceeds along similar lines, but the ions attached to Ca are more mobile than those attached to Fe or Al. The dilution appears to resemble a diffusion process. (22 references.) P. S. ARUP.

Aluminium ions in aluminium hydroxide, phosphate and soil-water systems. M. Raupach (*Nature, Lond.*, 1960, **188,** 1049—1050).—Linear curves of $-\log$(moles Al/l.) *vs.* pH for solutions in equilibrium with variscite $[Al(OH)_2,H_2PO_4]$ (**I**), clays and soils are shown and discussed in respect of similar results of other workers and of the possible existence of **I** in some acid soils. The solubility product and the slope of the curve in the system vary with the pH and the type of $Al(OH)_3$ present, hence either $[AlOH^{2+}][OH^-]^2$ or

$[Al_3(OH)_7^{2+}][OH^-]^2$ or $[Al(OH)_2^+][OH^-]$ can be constant. Theory and experiment confirm that min. solubility of **I** occurs between pH 4·2 and 5·2; reactions affecting the solubility of **I** under alkaline conditions are discussed. At all pH a knowledge of the ionic species present in soil phosphate solutions is critical for establishing the key reactions. W. J. BAKER.

Influence on plant growth of the breakdown of organic phosphorus compounds by micro-organisms. A. Szember (*Plant & Soil*, 1960, **13**, 147—158).—Micro-organisms capable of decomposing phytin (**I**) and lecithin (**II**) were isolated from soil by enrichment cultures. Inoculation of sterile sand cultures, containing **I** as the sole source of P, with some of these micro-organisms sometimes increased, but only slightly, and sometimes decreased the uptake of P by kale and radish. The availability of P from **II** was not affected by inoculation. There was evidence that both **I** and **II** could serve as sources of P for the plants even under sterile conditions. A. H. CORNFIELD.

Sulphur compounds in soil, their modification, relationship to microflora, and utilisation by plants. G. Simon-Sylvestre (*Ann. agron.*, 1960, **11A**, 309—328).—A review with 48 references. P. S. ARUP.

Effect of foliar-applied manganese on the concentration of manganese in oat roots.—E. Boken (*Physiol. Plant.*, 1960, **13**, 786—792).—Increase in the Mn concn. of oat roots one week after foliar sprays of $MnSO_4$ (2%) is not only maintained, but continues to increase strongly during the remainder of the growth period compared with basally fertilised crops. E. G. BRICKELL.

Availability of soil-chelated manganese to millet and its equilibria with other forms of manganese in the soil. J. M. Walker and S. A. Barber (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 485—488).—The uptake of Mn by millet from 12 Indiana soils, treated with lime, S and Mn, in pot tests was compared with the amounts of Mn extracted by chemical methods. Exchangeable Mn (extraction with N-NH_4OAc, pH 7) was correlated better with plant uptake of Mn than was "soil-chelated" Mn [extracted with 0·03N-$Zn(NO_3)_2$ after removal of exchangeable Mn]. The best prediction of Mn uptake was obtained from exchangeable Mn + (interaction between chelated Mn and % org. matter in soil). Somewhat lower correlations were obtained where air-dried than where moist soils were used for extraction. There was a highly significant negative correlation between soil pH and uptake of Mn by the plants. A. H. CORNFIELD.

Effect of ferrous sulphate, quinol and pyrolusite on yield and manganese uptake of oats on a sandy soil. E. Boken (*Plant & Soil*, 1960, **13**, 128—136).—Application of quinol (0·25 g. per kg. soil) to a Mn-deficient sandy soil (pH 7·7) in pot tests increased yields of dry matter and Mn uptake by oats to a greater extent than did application of $FeSO_4.7H_2O$. When the soil was also treated with ground pyrolusite (5 g. per kg. soil) the $FeSO_4$ was more effective than quinol in increasing the uptake of Mn by oats. A. H. CORNFIELD.

Effect of nitrogen carrier, nitrogen rate, zinc rate and soil pH on zinc uptake by sorghum, potatoes and sugar beets. L. C. Boawn, F. G. Viets, jun., C. L. Crawford and J. L. Nelson (*Soil Sci.*, 1960, **90**, 329—337).—On a non-calcareous sandy loam with a slightly alkaline reaction, N, 40—160 lb./acre, was supplied annually as $(NH_4)_2SO_4$, NH_4NO_3 or $Ca(NO_3)_2$ and $ZnSO_4$ was applied in one dressing at 0—16 lb./acre. With the heaviest rate of $Ca(NO_3)_2$, in some cases, lime and more Zn were used. A rotation of sorghum, potatoes, sugar beet and sorghum was established and leaves and soil were sampled at intervals. With both crops of sorghum the growth was greatest with $(NH_4)_2SO_4$ and least with $Ca(NO_3)_2$. Sugar beet gave the greatest growth with $Ca(NO_3)_2$ apparently benefiting from the residual N from the sorghum. Growth and yield of potatoes were unaffected by N source. Applications of Zn had no effect on yields or on the health of the plants. Zn uptake and concn. were greatest when $(NH_4)_2SO_4$ and least when $Ca(NO_3)_2$ was used. Application of Zn significantly increased Zn uptake and content. Zn uptake increased in most cases with soil pH and *vice versa*. Zn applied to the soil was slowly converted into forms not extractable by 0·1N-HCl. T. G. MORRIS.

Exchange of copper ions on mineral colloids. I. Adsorption phenomena. J. Muller (*Ann. agron.*, 1960, **11A**, 75—91).—The adsorption of Cu^{2+} (or of Cu^+ as a complex ion) by suspensions of Amberlite IR-120 (Ca form) or of several calcareous clays is in good agreement with the provisions of the Freundlich law. (21 references.) P. S. ARUP.

Chloride diffusion in soils as influenced by moisture content. L. K. Porter, W. D. Kemper, R. D. Jackson and B. A. Stewart (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 460—463).—Chloride diffusivities were measured in medium- and fine-textured soils at moisture tensions from 0·33 to 15 atm. Transmission factors obtained by dividing these values by diffusivity of Cl^- in water ranged from 0·310 to 0·027, depending on soil moisture content and texture. A. H. CORNFIELD.

Chemical nature of the nitrogen in the fulvic fraction of soil organic matter. F. J. Stevenson (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 472—477).—About 50% of the N occurring in the fulvic acid fraction of soil org. matter was present in compounds which were deaminated readily by acid and base hydrolysis. About 25% of the N existed as amino-acids and about 10% in amino-sugars. A selective adsorption technique was used for fractionating the fulvic extract of a brunizem soil into several components. A no. of these components were pigmented. All contained amino-acids and several contained amino-sugars. The N compounds which formed NH_3 by hydrolysis occurred primarily in association with pigments. The colloids of fulvic extracts were heterogeneous with respect to both charge and particle size. The pigments probably originated through the condensation of carbonyl compounds with amino-deriv. by mechanisms similar to those proposed for browning processes in natural products. A. H. CORNFIELD.

Subsidence and humification in peats. I. II. M. H. B. Hayes (*Dissert. Abstr.*, 1960, **21**, 759).—Paper electrophoretograms, differential thermal analysis thermograms, total acidity, C/N and org. matter/C ratios indicated that the dark surface horizon, the intermediate known horizon, and the black basal horizon of the soil were humified to a similar degree. The rates and extent of oxidation and solubilisation of tissue and peat were related to the lengths and to the moisture contents of the column. Org. matter in the drainage water was most concentrated in the short columns. O. M. WHITTON.

Decomposition and humification in soil of straw labelled with carbon-14. J. Szolnoki and É. T. Vágó (*Acta agron. Acad. Sci. hung.*, 1959, **9**, 407—414).—Most of the ^{14}C assimilated by barley plants (in the pre-waxy stage) placed in water containing $Ba^{14}CO_3$ and lactic acid, passes into the water-sol., constituents and the hemicelluloses. Mixtures of the straw with a calcareous sandy soil or a loam are found to have lost 82 and 86%, respectively, of their activity as $^{14}CO_2$ after incubation at 28° during 45 days. The humic acid fraction of the loam mixture shows greater, and the fulvic acid fraction much greater activity than the corresponding fractions in the sandy soil mixture. (25 references.) P. S. ARUP.

Incorporation of straw into soil: effects on soil microflora. G. Simon (*Ann. agron.*, 1960, **11A**, 5—53, 177—219).—In laboratory experiments, microbial activity is favourably stimulated, provided that ≮12 kg. of N (preferably NH_4^+-N or the org. N of lucerne) are applied per ton of straw. The N is at first fixed by the microflora, and later mineralised; the cellulose-consuming organisms are stimulated to produce a humus-like material from the straw. The validity of these findings is confirmed by pot experiments with barley. The beneficial effects increase with increasing soil-pH up to 7·3—7·5, and are most marked in well-aerated (not too heavy) soils, adequately supplied with moisture, P and K. (66 references.) P. S. ARUP.

Aspects of decomposition of cellulose in Canadian soils. I. Observations by microscope. II. Nitrate-nitrogen levels and carbon dioxide evolution. H. T. Tribe (*Canad. J. Microbiol.*, 1960, **6**, 309—316, 317—323).—I. Decomposition of cellulose in the soils examined was initiated by fungi, including *Rhizoctonia* sp. (dominant), *Humicola* and *Botryotrichum* spp. (sub-dominant). Subsequent breakdown was effected by bacteria and nematodes and the final stages by larger fauna, e.g., mites, collembolans, worms.

II. Decomposition of cellulose in a mull humus soil involved, initially, the uptake of NO_3^- leaving the soil deficient in NO_3^- for about 7 weeks. Some of this N was released later. Production of CO_2 from the decomposing cellulose was continuous. Soil fauna probably play a part in the mineralisation of N. A. G. POLLARD.

Vitamin B requirements of soil bacteria. J. Rivière (*Ann. agron.*, 1960, **11A**, 331—346).—A review and discussion with 48 references. P. S. ARUP.

Value of bush, grass or legume fallow in Ghana. K. Singh (*J. Sci. Fd Agric.*, 1961, **12**, 160—168).—Bush fallowing for 2 years was better than for 1 year; a 5- or 6-year period gave yields no better than did a continuous cropping system. When grasses and legumes were used as fallow cover crops, elephant grass followed by Guinea grass and pigeon pea were the most effective. Maize yields were increased after fallow on two southerly stations, but the reverse resulted on three northerly stations, the difference probably depending on type of fallow and availability of N. The pigeon pea, less demanding on soil N than are the grasses, gave good results on one station in Guinea Savannah. Burning the final cut of grass *in situ* increased the P, K and Mg status of the soil. (26 references.) E. M. J.

Effect of partial sterilisation of field soils by chemicals on differential microbial counts, carbon dioxide activity and rates of decomposition of organic matter. G. H. Elkan and W. E. C. Moore (*Canad. J. Microbiol.*, 1960, **6**, 329—347).—Use of $CaOCl_2$, $HgCl_2$ and Na propionate as soil-sterilising agents usually caused an initial increase

in respiration rates especially in the case of the propionate which diminished fungal and increased bacterial no. in the soil. With all treatments CO_2 production was lowered within 4 weeks; this after-effect persisted for about 4 months (13 months in the case of $HgCl_2$, 100 lb./acre) after treatment, when the lowered levels of micro-organisms and CO_2 production were still marked. No consistent relationships were apparent between microbial no. or respiration rates and changes in org. matter contents of the soils.
A. G. POLLARD.

Yields and composition of five grasses in the humid mountains of Puerto Rico as affected by nitrogen fertilisation, season and harvest procedures. R. Caro-Costas, J. Vicente-Chandler and J. Figarella (*J. Agric. Puerto Rico*, 1960, **44**, 107—120).—Molasses grass gave lower yields of dry matter than did Napier, Guinea, Para and Pangola grasses. Yields of the last four were very similar, as were responses to N fertilisers and seasonal growth rates with cutting or simulated grazing management; the only exception was that Guinea grass outyielded the others with the simulated grazing management. The lowest yields occurred during Dec.–March. Seasonal variations were accentuated by N fertilisation. Molasses grass responded no better with 400 lb. than with 200 lb. of N per acre, but yields of the other grasses increased with rate of application of N up to 400 lb. per acre. Protein content of all grasses was similar, increased with N rates, was higher during seasons of slow growth, and lower with cutting than with simulated grazing. There was little difference in efficiency of N utilisation by the four high-yielding grasses. Efficiency was lower with grazing management, higher during seasons of fast growth, and decreased with increasing rate of applied N. A. H. CORNFIELD

Protein from sugar cane. D. H. Parish (*Nature, Lond.*, 1960, **188**, 601).—Protein (6% by dry wt.) in sugar-cane leaves can be extracted by maceration, filtration and heat-coagulation; heat-coagulated protein (**I**) from sugar manufacture has ~1% N, but ppt. having up to 8% N can be prepared from cane juice in the laboratory. Protein-cake from both sources serves as soil fertiliser in N-deficient areas. Mauritius produces $\sim 10^5$ tons of **I** annually.
W. J. BAKER.

Mineralisation and utilisation of nitrogenous reserves of soil. G. Lefèvre and G. Hiroux (*Ann. agron.*, 1960, **11A**, 135—162).—Previously described calculations (cf. *ibid.*, 1956, **7**, 23) are applied to observations on plots variously treated with farmyard manure or mineral N. "Premineralisation" (by which org. N reserves are converted into readily mineralisable matter) is brought into play by a biological equilibrating process which is stimulated by losses in mineral N due to leaching or to utilisation by plants. A reversal (conversion of mineral N into the premineralised stage) may occur on the application of mineral N or through non-utilisation of available N. The application of farmyard manure, especially if accompanied by mineral N, tends to delay mineralisation and to build up reserves which become available during the second or third year after application. P. S. ARUP.

Sequential products of anaerobic denitrification in Norfolk soil. F. B. Cady and W. V. Bartholomew (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 477—482).—Nitric oxide was the first gaseous denitrification product to appear during incubation of a sandy loam, to which ^{15}N-labelled NO_3^- had been added, under anaerobic conditions in a closed circulating system. NO_2^-, also derived from added NO_3^-, was associated with NO, which presumably was derived from decomposition of HNO_2. NO gradually decomposed and its disappearance was accompanied by the appearance of and increase in N_2O. N_2 then appeared and after complete disappearance of N_2O the amount of N_2 accounted for 83—95% of the N added as labelled NO_3^-. A. H. CORNFIELD.

Utilisation of nitric oxide by micro-organisms and higher plants. C. A. Fewson and D. J. D. Nicholas (*Nature, Lond.*, 1960, **188**, 794—796).—Nitric oxide reductase is a metallo flavo-protein with max. activity at pH 8. It is inhibited by Fe and Ca chelating compounds, e.g., $\alpha\alpha'$-dipyridyl and by Fe deficiency. Micro-organisms and probably higher plants can assimilate NO only when grown in NO_3^- media and not with NH_4^+-N. Results suggest that NO is intermediate in assimilatory and dissimilatory NO_3^- reduction and in hydroxylamine nitrification. (29 references.) R. J. M.

Sampling of soil for determination of nitrogen. R. Vanstallen (*Agricultura*, 1960, **8**, 247—258).—Representative composite samples giving the % of N at a probability level of 10% can be obtained from 40 bores dispersed uniformly over areas up to 50 acres having no recent manuring or 60 bores on recently manured land. The no. of bores necessary is not influenced by the nature of the surface.
P. S. ARUP.

Plate method for studying breakdown of synthetic and natural silicates by soil bacteria. D. M. Webley, R. B. Duff and W. A. Mitchell (*Nature, Lond.*, 1960, **188**, 766—767).—Insol. silicates are ground in a ball mill and directly incorporated into molten agar as described for insol., phosphates (Louw and Webley, *Nature, Lond.*, 1958, **182**, 1317). E. G. BRICKELL.

Fertiliser evaluation. IV. Use of ^{32}P-labelled fertilisers. D. R. Bouldin and C. A. Black (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 491—496).—Definitions for yields of fertiliser P in crops by conventional and isotopic techniques are proposed. Experimental results showed that these two kinds of yields of P and the fertiliser evaluations derived from them are not necessarily identical. Explanations for the differences are advanced. A. H. CORNFIELD.

Leaf and soil analyses as a means of guiding the fertilisation of citrus fruit plantations. D. H. Chapman (*Fruits d'outre mer*, 1960, **18**, 435—441).—The value of leaf analysis (alkali and alkaline earth metals, N, S, P and Cl values and trace elements: B, Co, Fe and Mn) and soil analysis (alkali and alkaline earth metals and phosphates) in assessing fertiliser requirements for optimum yield and quality of fruit is discussed. Data on two orange orchards are presented.
J. V. RUSSO.

Experiments in small pots for testing efficiency of organic fertilisers. R. Chaminade (*Ann. agron.*, 1960, **11A**, 121—133).—Rye-grass is sown in 1-litre pots (1000 seeds per pot) containing the prepared soils, and cropped at intervals in order to determine the productivities in dry matter resulting from the various treatments.
P. S. ARUP.

Economical long-term utilisation of organic fertilisers in sandy soils. S. Egerszegi (*Acta agron. Acad. Sci. hung.*, 1959, **9**, 318—340).—Long-term satisfactory results as regards fertility, water-conservation and activity of the soil microflora have been obtained by the deposition of farmyard manure in layers at 35 and/or 60 cm. depth. The directions given stress the importance of leaving these layers undisturbed and of restricting cultivation to loosening rather than turning the soil. (10 references.) P. S. ARUP.

Nitrogen studies in a coffee soil. II. Influence of mulch on natural and fertiliser levels of nitrate and ammonia in the top-soil. III. Comparative efficiency of ammonium sulphate and urea fertilisers in presence and absence of an organic mulch as measured in terms of crop yields. J. B. D. Robinson (*J. agric. Sci.*, 1961, **56**, 49—59, 61—64).—II. Surface mulching reduces NO_3^--N levels following N fertiliser application compared with an unmulched soil. Natural NO_3^--N levels are lower under a gravel mulch than under a Napier grass mulch. Whilst under a mulch there is little difference between the effects of $NaNO_3$ and $(NH_4)_2SO_4$, the former raises NO_3^--N levels in bare soils more than does the latter. Urea and $NaNO_3$ have similar effects on NO_3^--N levels in a wet soil. With a partial mulch cover efficiency is greatest where the N fertiliser is applied to the unmulched areas. In long rainy periods split N applications are more efficient.

III. A single application of $(NH_4)_2SO_4$ is more efficient than one of urea in absence of an org. mulch. In the presence of a complete mulch cover split applications are more beneficial than single, the reverse holding on bare soils. Splitting of applications is less important with $(NH_4)_2SO_4$ than with urea. M. LONG.

Soluble aluminium as factor in soil acidity and in response of plants to lime. W. R. Hourigan (*Dissert. Abstr.*, 1960, **21**, 719—720).—In sand cultures, the growth of barley is reduced by 11, 6 and 300 p.p.m. of Al, Mn and Fe, respectively, but in soils of various pH, toxic effects are observed for Al only. In liming experiments with ^{45}Ca as tracer, previously limed soils at pH 5—7 give greater yields of barley and lucerne than do freshly limed soils, but the uptake of Ca is greater from the latter than from the former. The best yield-responses by barley are obtained at pH 6 for the topsoil, and by lucerne at pH 7. In titrations of buffers or of buffers + soil, with N-HCl and with N-$AlCl_3$, the Shoemaker, McLean and Pratt (SMP) buffer is probably more efficient in measuring the acidity due to $AlCl_3$ than are the Woodruff and Mehlich buffers. Comparative experiments in the titration of leached and untreated soils are also described.
P. S. ARUP.

Lime and fertiliser incorporation for lucerne production. J. R. Love, A. E. Peterson and L. E. Englebert (*Trans. Wis. Acad. Sci. Arts Lett.*, 1960, **49**, 161—169).—The effects of applying lime + fertiliser before and after ploughing compared with those where each material was applied separately either before or after ploughing, on lucerne–brome hay, were studied. Regardless of method of incorporation, the K content of the top 2 in. was 2—4 times higher than that in any succeeding 2-in. depth of the plough layer. P distribution was similar to that of lime. No differences, due to method of lime and fertiliser incorporation, were observed in stand counts of lucerne taken in the spring of the second hay year, chemical analyses of lucerne in the first two years or in total yields of lucerne–brome hay for 3 years. (11 references.) E. M. J.

Efficiency of various particle-size fractions of limestone. H. L. Motto and S. W. Melsted (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**,

488—490).—The changes in pH with time of three acid soils after application of agricultural limestone of varying particle sizes in greenhouse tests was related to time, rate of application, particle size and soil type. Equations relating these variables are presented. Limestone particles of 10 to 28-mesh size were about 14% as effective as those in the <100-mesh fraction, whilst particles >10-mesh were of little value. For practical purposes most limestones should be ground to pass a 40-mesh sieve. A. H. CORNFIELD.

Use of various indicators for complexometric determination of calcium and magnesium in fertilisers, soils and plants. E. Jouis and M. T. Lecacheux (*Ann. agron.*, 1960, **11A,** 113—120).—The use of seven indicators is investigated. For the titration of Ca with 0·01M-EDTA at pH 12, murexide + 2-Naphthol-green is recommended, and for Ca + Mg (after the removal of Fe and Mn by means of diethyldithiocarbamate), Eriochrome-black T. Working directions are given. P. S. ARUP.

Plant Physiology, Nutrition and Biochemistry

Physical aspects of plant growth. M. B. Russell (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 439—440).—An address. A. H. CORNFIELD.

Action spectra of light-saturated photosynthesis. G. C. McLeod (*Plant Physiol.*, 1961, **36,** 114—117).—In saturating light the rate of photosynthesis is dependent on the wavelength of the incident light. The Emerson enhancement effect may be responsible for the structure of the high-intensity action spectrum. E. G. BRICKELL.

Cyanide and chelating agent effects on *in vitro* CO_2 fixation in sweet orange leaves. A. Wallace and R. T. Mueller (*Plant Physiol.*, 1961, **36,** 118—120).—The major effect of CN^- was the stabilisation of oxaloacetic acid by formation of a cyanohydrin. Removing heavy metals did not alter the increased fixation resulting from CN^- and the latter overcame the inhibitory effects of heavy metals added to the phosphoenol pyruvate reaction system except for Zn. The stimulating effect of a chelating agent was additive with either CN^- or N_3^-. E. G. BRICKELL.

Dependence of anthocyanin synthesis on light. R. Kandeler (*Flora*, 1960, **149,** 487—519).—The dependence holds good under conditions where heating effects are excluded. Published data reveals the agency of at least five distinct light-reactions. The inhibiting effect of white light on the synthesis is shown to be distinct from that concerned with chlorophyll synthesis. The stimulating effect of high-energy dark-red on anthocyanin synthesis in red cabbage can be replaced partly by additions of sucrose, adenosinetriphosphoric acid or ascorbic acid, but not by NaOAc, phloroglucinol or L-phenylalanine. The probable mechanism of the dark-red effect is considered. (120 references.) P. S. ARUP.

Co-operation of various factors in decoloration of anthocyanins in begonias. H.-L. Jürgensmeier (*Planta*, 1961, **56,** 233—235).—A marked inverse relationship is observed between the production of anthocyanin and that of oxalic acid–oxidase activity in the growing plants. (10 references.) P. S. ARUP.

Mineral salt deficiencies and secondary carotenoids in green algae. G. Dersch (*Flora*, 1960, **149,** 566—603).—Deficiency of N, P, S, K or Fe (but not of Mg or Mn) causes *Ankistrodesmus* to produce the secondary carotenoid astaxanthine; this effect is independent of illumination or of the concomitant reduction in the synthesis of chlorophyll and the normal carotenoids; the astaxanthine disappears after the restoration of normal conditions. Diphenylamine inhibits the synthesis of astaxanthine, but not that of fat; phenol retards both processes. Deficiency of Fe specifically inhibits the synthesis of carotene. (67 references.) P. S. ARUP.

Reversible photochemical reduction reactions of chlorophyll and its analogues and derivatives. A. A. Krasnovskiĭ (*Usp. Khim.*, 1960, **29,** 736—759).—Reaction mechanisms are discussed. (112 references.) A. L. B.

Endogenous rhythm and carbon dioxide metabolism in plants with diurnal acid rhythm. E. L. Nuernbergk (*Planta*, 1961, **56,** 28—70).—The occurrence or absence of the De Saussure effect (nightly assimilation of CO_2 through malate) in the leaves of succulent plants is not connected with noticeable morphological differences, but diurnal variations in the sap-pH are generally much greater in plants which show the effect than in those which do not. A 24-h. rhythmic effect is discernible in *Bryophyllum daigremontianum* in spite of the imposition of considerable photoperiodic variations; an annual rhythm can also be detected through seasonal variations in responses to artificial photoperiodic changes. Flower initiation has no effect on the course of CO_2-assimilation. The mechanism of photoperiodic CO_2 assimilation is considered on the basis of these and other observations. (42 references.) P. S. ARUP.

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Formation of the glucose derivative of 3-amino-1,2,4-triazole [3AT] under physiological conditions. J. F. Fredrick and A. C. Gentile (*Physiol. Plant.*, 1960, **13,** 761—765).—The glucose derivative of 3AT is formed by an interaction between 3AT and the Cori ester. E. G. BRICKELL.

Occurrence and detection of purine and pyrimidine compounds in *Zea mays* seedlings, and experiments in purine synthesis. H. Gräser (*Flora*, 1960, **149,** 520—542).—The presence of several nucleotides and ribosides is demonstrated by paper-chromatography in the coleoptile and primary root; the adenylic acid system is identified by electrophoretic analysis. The relative proportions of purine to pyrimidine deriv. vary with age, and also as between the coleoptile and root. The feeding of seedlings with glycine, glutamic acid, aspartic acid or (in low concn.) formamide results in increased production of adenylic acid and inosine in the coleoptile. With higher concn. of formamide the effect is reversed. P. S. ARUP.

Effects of calcium, potassium and magnesium on oxalic, malic and citric acid content of Valencia orange leaf tissue. G. K. Rasmussen and P. F. Smith (*Plant Physiol.*, 1961, **36,** 99—101).—Oxalic acid in the tissue was increased by Ca in the presence of K and by K in the presence of Ca. Leaf-malic acid was positively correlated with -Ca but not influenced by -K. Leaf-citric acid was unaffected by the three cations. Total water-sol., org. acids were affected only by Ca. Mg as a reciprocating cation showed opposite effects from those of both K and Ca. E. G. BRICKELL.

Cation-exchange capacity and pectin gradients in leek root segments. W. M. Crooke, A. H. Knight and I. R. MacDonald (*Plant & Soil*, 1960, **13,** 123—127).—Cation-exchange capacity, pectin and total N contents and respiration rate of segments of leek roots were highest at the root tip and decreased with increasing distance from the tip (up to 14 cm.). A. H. CORNFIELD.

Effect of root to solution ratio in ion absorption experiments. L. Jacobson, R. J. Hannapel, M. Schaedle and D. P. Moore (*Plant Physiol.*, 1961, **36,** 62—65).—Large differences in absorption behaviour are caused by changing the ratio wt. of root/vol. of solution, the liberation of Ca and other absorption-modifying substances from the roots being the most important factor. Changes in pH as a result of absorption activity cause a reduction in absorption rates but this factor may be eliminated by appropriate control measures. E. G. BRICKELL.

Influence of calcium on selectivity of ion absorption process. L. Jacobson, R. J. Hannapel, D. P. Moore and M. Schaedle (*Plant Physiol.*, 1961, **36,** 58—61).—Ca drastically alters the ratio of absorbance of Na and K from a mixture of the two and this controlling behaviour was found in the roots of six different species of plants. E. G. BRICKELL.

Uptake of calcium by excised barley roots. D. P. Moore, L. Jacobson and R. Overstreet (*Plant Physiol.*, 1961, **36,** 53—57).—Uptake is largely non-metabolic. Much of the Ca in young barley roots is associated with the cell surface region and this is active in influencing the absorption of other ions. E. G. BRICKELL.

Effect of various cations upon absorption of carrier-free caesium. R. Handley and R. Overstreet (*Plant Physiol.*, 1961, **36,** 66—69).—In concn. 0—0·10 mequiv. per l. Na, Li, Ca, Ba and Mg had essentially no depressant effect upon the uptake of ^{137}Cs whereas K, Rb, NH_4 and Cs were markedly effective. At higher concn. all the ions investigated inhibited the absorption of ^{137}Cs. E. G. BRICKELL.

Effect of sodium bicarbonate on iron absorption by orange seedlings. E. F. Wallihan (*Plant Physiol.*, 1961, **36,** 52—53).—*Citrus sinensis* is sensitive to $NaHCO_3$ and the resulting reduction of Fe absorption by the roots follows immediately upon exposure to the bicarbonate system. E. G. BRICKELL.

Effect of phosphorus and pH on iron chlorosis of the blueberry in water culture. R. S. Holmes (*Soil Sci.*, 1960, **90,** 374—379).—Rooted cuttings of blueberry were grown in aerated solution culture, either at constant pH 4 with varying P levels (0—60 p.p.m.) (Group 1) or at a constant P level (20 p.p.m.) and at pH 4—8 (Group 2). Ca, Mg, N, K and S were added and minor elements Mn, Mo, B, Zn and Cu, and Fe (5 p.p.m.) as $FeCl_3$. After 4 months, group 1 plants differed less widely in growth in relation to treatment than did those of group 2. Group 1 plants grown with the intermediate P levels were the same size and shape but those with the lowest and highest P levels were relatively smaller. P deficiency was evident with the lowest and chlorosis with the highest P level. Group 2 plants were more variable in size than those in group 1; at pH 4 and 5 plants were similar to those in group 1, but at higher pH levels the plants decreased in size with increase in pH. Chlorosis also increased with pH in these plants. At pH 8 little or no growth occurred. When the $FeCl_3$ in the solution at pH 7 was replaced by FeEDDHA the chlorosis disappeared and in 3 months the plants had

more than doubled in size. The P and Fe contents of the plants varied with the P and Fe levels of the solution. High P levels lowered the Fe level in the nutrient thereby inducing chlorosis in the plants, notably in young leaves. Increasing pH tended to decrease leaf-Fe levels especially in young plants. T. G. MORRIS.

Distribution of boron in *Oenothera* species. Histochemistry of plants. I. H. O. Glek and W. Wagner (*Ber. dtsch. bot. Ges.*, 1960, **73**, 463—470).—The highest concn. of B are found in the pistils, the vegetative growing points, and the leaves, and the lowest in the pollen, fruits and seeds. Accumulations of B amount (on dry basis) to 10—20 times the concn. found in the soil. The concn. decrease via the stamens to the pollen, but increase via the pistils to the stigmata; the concn. in the pollen are 8—9% (on dry basis) or ~50% of those found in the stigmata. Fertilisation is not accompanied by extra transport of B to the stigmata. The observed optimum concn. of B for the growth of the pollen tubes *in vivo* correspond with those found *in vitro*. (13 references.) P. S. ARUP.

Migrations of mineral substances and development of ionic composition in growing tomato plants. J. R. Ansiaux (*Ann. Physiol. veg. Brussels*, 1960, **5**, 19—260).—Comparisons between sterilised and fruiting plants reveal a competition between the fruits and the vegetative parts resulting in the gradual accumulation of NO_3^-, PO_4^{3-} and K^+ in the fruits; these ions constitute >90% of the mineral matter in the ripe fruits. A difference is observed between the patterns of mineral composition of the stems and leaves; the dry matter and fixed N content of the leaves decrease with age, whilst the leaf-Mg increases. The above effects are not modified by alterations in climatic or nutritional conditions. The effects of the omission of any two of the ions K^+, Ca^{2+} or Mg^{2+} from the nutrient solution are examined. P. S. ARUP.

Translocation from leaves of rye. A. Mayer and H. K. Porter (*Nature, Lond.*, 1960, **188**, 921—922).—Single leaves of a rye plant were supplied with $^{14}CO_2$ and the distribution of radioactivity throughout the tiller determined after a period in light and air. No activity was found in leaves other than those receiving $^{14}CO_2$ and little in other parts of the tiller. Of the activity of eluates from charcoal column chromatography of ethanol/water extracts of the tiller 81% was in sucrose, 13% in hexoses and the rest in tri- and tetra-saccharides. S. A. BROOKS.

Comparative study of carbohydrate translocation in apple, raspberry and soya-bean. J. W. A. Burley (*Dissert. Abstr.*, 1960, **21**, 737).—Experiments with ^{14}C as tracer show, for raspberry and soya-bean, radiochemical equivalence of glucose and fructose in the phloem tissue, and increases in the ratio of radioactive sucrose to hexoses with the distance of translocation; the theory of carbohydrate transport as sucrose is thus supported. In apple, fructose-radioactivity is comparatively slight; the evidence points to oligosaccharide-transport by raffinose and stachyose; the galactose eventually derived from these sugars is presumably transformed into glucose immediately on liberation. P. S. ARUP.

Sorption of fluorine by citrus foliage from equivalent solutions of [fluorine compounds]. R. F. Brewer, F. H. Sutherland and F. B. Guillemet (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 214—219).—Navel orange trees sprayed five times with solutions of HF, NaF, NH_4F or H_2SiF_6, 0·0004—0·10N or with CaF_2 at the lowest concn. generally absorbed F most readily from the HF and NaF sprays. L. G. G. WARNE.

Effect of atmospheric fluoride on respiration of bush beans. H. G. Applegate and D. F. Adams (*Bot. Gaz.*, 1960, **121**, 223—227).—In concn. below those causing visible symptoms of fluorosis, atm. F^- increased the O_2 intake of the bean plants. Data for the uptake of F by the tissue are recorded. A. G. POLLARD.

Effects of hydrogen fluoride gas on bearing navel orange trees. R. F. Brewer, F. H. Sutherland, F. B. Guillemet and R. K. Creveling (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 208—214).—Navel orange trees were adversely affected by HF in the air to the extent of 2—3 p.p.b. (10^9). At this low concn. growth was reduced but no characteristic symptoms of damage occurred. At higher concn. leaf chlorosis developed, associated with F concn. in the leaf of 75 p.p.m. or higher on a dry wt. basis. L. G. G. WARNE.

Composition of nectar and mechanism of its secretion. I. U. Lüttge (*Planta*, 1961, **56**, 189—212).—Published data are reviewed, and new information is contributed concerning the nectar of a no. of plants, including the amino-acid, protein, and org. and inorg. phosphate content of the nectar of *Musa sapientum* L. Allantoin and allantoic acid are found in maple nectar, a tyrosinase occurs in the nectar of *Lathreae clandestina*, and uridine diphosphate-glucose in *Abutilon* nectaries. Resorption of $^{32}PO_4^{3-}$ from the nectar to the nectary tissue is demonstrated in several species. (55 references.). P. S. ARUP.

Ribonucleic acid and flower initiation. D. Hess (*Planta*, 1961, **56**, 229—232).—On the basis of published observations, a theory is proposed which postulates the operation of two competitive ribonucleic acid mechanisms, viz., a reproductive and a vegetative mechanism, the latter being inactivated under conditions favouring flower initiation. P. S. ARUP.

Determination of fructosans in presence of lower sugars in biological material. J. P. J. Casier and L. Sue (*Agricultura*, 1960, **8**, 269—298).—This method is applicable to material containing fructosans in appreciably larger amounts than other saccharides easily hydrolysed by 0·02N-HCl at the b.p. within 15 min. The dry sample is extracted under reflux, first with anhyd. ether, then (for the complete removal of the lower sugars) with a mixture of EtOH, Pr^nOH, Bu^nOH and water (80 : 15 : 5 : 1, by vol., 5 portions), and finally with warm water (several portions). If much inulin is present, the extraction vessel (a cylindrical separating-funnel) must be surrounded by a heating-mantle in order to keep the inulin in solution. A suitable aliquot of the filtered (through glass wool) combined (well mixed) aq. extracts is hydrolysed with 0·02N-HCl at the b.p. during 20 min.; the fructosans are then evaluated by a chromatographic or reductimetric determination of the products of hydrolysis. No destruction of fructosans occurs during any of the stages of the process. (27 references.) P. S. ARUP.

Further studies of synergism in slit pea test. B. E. Michel (*Plant Physiol.*, 1961, **36**, 92—98).—Indol-3-ylacetonitrile (IAN), 1-naphthylacetonitrile (NAN), cyclohexylacetic acid (CHAA), and possibly γ-phenyl-n-butyric acid (PBA) synergise with indol-3-ylacetic acid (IAA) and 1-naphthylacetic acid (NAA), but not with 2,4-D in the slit pea curvature test. Skatole and indole show no synergic effects. Certain concn. of IAA, PBA, and NAN alone, are active in this test. The first two decrease negative curvature; the last increases it. The interpretation of the slit test is more complex than that of the cylinder test. The slit internode curves in response to auxins, but curvature is no guarantee of auxin activity. E. G. BRICKELL.

Naturally occurring growth substances. II. An improved straight growth test and its applications. D. G. Crosby, R. V. Berthold and R. Spenser, jun. (*Plant Physiol.*, 1961, **36**, 48—51).—A very satisfactory first internode test for oat varieties is described, with the Forkedeer variety showing sections of outstanding uniformity, sensitivity, range of effective concn. and pH, and availability. Determination of hormone profiles of aq. and ethereal extracts of the Alaska pea, based on the Forkedeer bioassay, failed to provide conclusive evidence of IAA in this variety. Several other unidentified growth stimulants, insol. in ether, were detected. E. G. BRICKELL.

Influence of auxins on salt and water uptake. G. Swenson and H. Burström (*Physiol. Plant.*, 1960, **13**, 846—854).—Auxins increase the uptake of cations in the order K > Na > Mg > Ca. Water uptake is also reduced but in certain instances much less than that of ions. Prevention of root growth by decapitation does not annihilate the auxin effect. E. G. BRICKELL.

***In vitro* destruction of auxin-labelled with ^{14}C.** P. E. Pilet (*Physiol. Plant.*, 1960, **13**, 766—775).—IAA labelled with ^{14}C is rapidly destroyed under the action of an auxin-oxidase system in two distinct stages, (i) shortening of the side chain with formation of indole-3-aldehyde and (ii) degradation of the nucleus (pyrrolic decyclisation) with the formation of two products, not indolic compounds, either directly from IAA or, more probably, at the cost of (i). E. G. BRICKELL.

Enzymic production of growth substances from tryptophan under influence of a native inhibitor. E. Libbert (*Planta*, 1961, **56**, 1—22).—The production of the following substances from tryptophan by a crude extract of green pea plants is demonstrated by paper-chromatography: indol-3-ylacetic acid (IAA), indol-3-ylcarboxylic acid (ICA), indole-3-aldehyde (IA), tryptamine (TNH_2), and two unknown indolyl deriv. of which one (Xa) is established as the source of ICA and IA. The isolated Xa can, however, be made to yield IAA through the agency of the enzyme, but this process can be blocked by the presence of a non-acidic Et_2O-sol. inhibiting enzyme which can be obtained from the plant extract; this enzyme increases the production of Xa, and consequently that of ICA and IA. (48 references.) P. S. ARUP.

Effect of indolylbutyric acid on respiration and nitrogen metabolism in Marianna 2624 plum softwood stem cuttings. D. K. Strydom and H. T. Hartmann (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 124—133).—Cuttings, whose basal ends had been dipped in indolylbutyric acid showed in the basal portion a higher respiration rate than did the controls and by the time roots appeared the rate was four times that of the untreated cuttings. The leaves and the basal portion of the treated cuttings had an increased and the median region a decreased content of N. An increase in free amino-acids

in the treated cuttings was apparent after 48 h. and the value continued to increase until roots appeared. L. G. G. WARNE.

Separation and study of the neutral fraction of growth-substances in immature maize grains. G. Beauchesne and J.-P. Jouanneau (*C. R. Acad. Sci., Paris,* 1960, **251,** 2396—2398).—Previous procedure (*ibid.,* 1957, **244,** 112) has been improved by filtering the 50% ethanolic extract of maize through an intimate mixture of ion-exchange resins (IR 120 + Dowex 2) whereby the pH remains constant and an effective separation into four growth-active fractions can be achieved directly. The filtrate is the neutral fraction, containing reducing sugars, indole compounds, purines and pyrimidines. Selective elution of the mixed-bed with 2N-aq. NH_3 and saturated aq. $(NH_4)_2CO_3$ yields three fractions: (i) bases, basic amino-acids and purine-bases, (ii) neutral and acidic amino-acids, and (iii) indole-acids and acids of purine bases. All four fractions promote cellular elongation in the mesocotyl test; neutral and basic fractions both show strong cell-division activity. W. J. BAKER.

Relation of chemical structure to plant growth-regulator activity in the pineapple plant. II. Compounds related to phenoxyalkylcarboxylic acids, phthalamic acids and benzoic acids. D. F. Gowing and R. W. Leeper (*Bot. Gaz.,* 1960, **21,** 249—257).—Tests with 151 substances are recorded. Induction of flowering (forcing) of pineapple was generally effected by alkyl and terpenoid esters of 4-chloro-, 2,4-dichloro-, and 2,4,5-trichloro-phenoxyalkyl carboxylic acids. High concn. of some of these compounds were toxic or inhibitory. Ring-substituted phenylmercaptoacetic acids and substituted phenylglycines had little or no forcing action. Forcing of pineapple and positive responses in the pea test were associated in DL- and L-non-aromatic amino-acid deriv. of the substituted phenoxyacetic acids. The D-amino-acid deriv. forced pineapple but had substantially no activity in the pea test. Tryptophan derivatives of 2,4-dichloro- and 2-methyl-4-chloro-phenoxy-4-butyric acids had no forcing action although phenylalanine derivatives were active. Among 2,4-dichlorophenylalkyl ethers examined the n-hexyl, n-decyl and n-tetradecyl, 2- chloroethyl and 2-methoxyethyl ethers caused forcing but the n-propyl, methyl and methyl ethers were not active. Of various derivatives of benzoic, salicylic, phthalic and phthalamic acids tested only 2,3,6-trichlorobenzoic acid showed forcing effects. A. G. POLLARD.

Correlation between inactivation of 2,4-D and cessation of callus growth in bean stem sections. M. K. Bach and J. Fellig (*Plant Physiol.,* 1961, **36,** 89—91).—Radioactivity from ^{14}C-carboxyl-labelled 2,4-D largely remained in the tissue in an alcohol-sol. form chromatographically distinct from 2,4-D. Only relatively small amounts of radioactivity were found in CO_2 or released into the medium. Cessation of growth is therefore linked with the disappearance of free 2,4-D from the sections. E. G. BRICKELL.

Effects of gibberellic acid, indolylacetic acid [IAA], coumarin and perloline on perennial rye-grass (*Lolium perenne,* L.). S. O. Fejer (*N.Z. J. agric. Res.,* 1960, **3,** 734—743).—Gibberellic acid (**I**) produced initial elongation resulting in taller plants, followed by total reduction of plant size caused by reduced rate of tillering. At the rates used flowering was not promoted and green wt. of inbred lines treated with **I** showed no response. Coumarin had little effect on growth or tillering. IAA promoted the rooting of cuttings. Perloline had a growth-inhibiting effect. Calculated data are given on a no. of significant interactions between individual chemicals or their rates of application with the plant material. (18 references.) E. M. J.

Action of gibberellic acid in the *Avena* coleoptile curvature test. S. Saebø (*Physiol. Plant.,* 1960, **13,** 839—845).—Gibberellic acid is active in the *Avena* coleoptile curvature test, increasing negative and decreasing positive curvatures by a few degrees. An interaction between GA and IAA was noted. E. G. BRICKELL.

Effect of light and gibberellic acid on internode growth in *Pisum sativum.* P. J. M. Sale and D. Vince (*Physiol. Plant.,* 1960, **13,** 664—673).—Inhibition of internode elongation by light was partly but not completely prevented by the application of large doses of gibberellic acid (GA). Internode lengths of plants in light treatments plus GA were in all cases less than those grown in the dark plus GA. In dwarf varieties the shortest internodes occurred always in red light; in tall, the relative effectiveness of red and blue light varied with the duration of irradiation. When GA was applied the different responses of tall and dwarf varieties to red and blue light were almost completely eliminated. There was no interaction between GA and infra-red; both caused internode elongation, and they were almost completely additive in their effects. E. G. BRICKELL.

Effects of gibberellins and dinitrophenol on respiration of stem tips of a long and a short pea variety. A. Goffeau and K. Buffel (*Agricultura,* 1960, **8,** 299—318).—After 9 days, gibberellin (at 3 p.p.m. in the nutrient solution) increased the stem length by ~80% in the long, and 100% in the short variety; the respiration of the (1-cm.) tips was not affected in the long, but decreased by ~29% in the short variety; the overall respiration of the stems was increased. At 5×10^{-5}M, DNP increased respiration in the tips of the long, but not in the short variety. Negative results were obtained with glucose and arsenate. The significance of these findings is considered with respect to effects on the mechanisms of respiration and of energy expenditure. (24 references.) P. S. ARUP.

Influence of growth stage on the response of red clover, *Trifolium pratense,* L., to gibberellic acid. J. L. Stoddart (*Ann. appl. Biol.,* 1960, **48,** 800—810).—Red clover seedlings sprayed with gibberellic acid (GA, 0·0001 g. per plant in 10 ml. spray) before the seventh leaf stage developed into single-stemmed plants. Treatment at the third- and fourth-tiller stage had no effect on final no. of stems; emergence was earliest and the no. of heads per plant greatest when treatment was delayed until this stage. In the first harvest year significant differences in the no. of heads per stem were obtained with certain treatments, particularly those which had two applications of GA (0·0005 g. per plant) during elongation of the first four internodes. A. H. CORNFIELD.

Breaking the rest period in blackcurrant with gibberellic acid and low temperature. I. Modlibowska (*Ann. appl. Biol.,* 1960, **48,** 811—816).—Application of gibberellic acid (GA) (100 p.p.m.) in Sept., Oct. or Nov. to blackcurrant plants which were then brought into the greenhouse broke the rest period of the plants. Chilling (2°) the plants for 2 weeks was ineffective in breaking the rest period. The highest % bud burst was obtained when GA was applied immediately after defoliation. Delay in application of GA after defoliation decreased and delayed plant response because of poor penetration of GA. Very early application or application of excessive amounts of GA caused shedding of undeveloped buds. A. H. CORNFIELD.

Sustained treatment with gibberellic acid of maize plants carrying one of the dominant genes Teopod and corn-grass. N. H. Nickerson (*Amer. J. Bot.,* 1960, **47,** 809—815).—Repeated applications (every 3 days from seedling to tasselling stages) of gibberellic acid to the mutants, induced modifications which rendered the plants substantially normal in phenotype. A. G. POLLARD.

Inhibition of transport of gibberellic acid (GA) in hypocotyl segments by tri-iodobenzoic acid; new agar-block test for gibberellin. T. Kentzer and E. Libbert (*Planta,* 1961, **56,** 23—27).—The inhibition is demonstrated in sunflower seedling segments which have previously been treated with TIBA. The level of transport is determined by means of agar acceptor-blocks for GA; the blocks are subsequently attached to the surfaces of test plants (pea seedlings) from which the epidermis has been removed. P. S. ARUP.

Inhibition of root development on petioles and hypocotyls of dwarf bean (*Phaseolus vulgaris*) by kinetin. E. C. Humphries (*Physiol. Plant.,* 1960, **13,** 659—663).—Kinetin inhibits root formation on petioles and hypocotyls. It induces callus on the former but not on the latter. When kinetin is subsequently withdrawn normal roots originate from the callus. Kinetin counteracts the effect of NAA in inducing root formation. E. G. BRICKELL.

Action of hexachlorophene on plant roots. A. G. Norman (*Antibiotics & Chemotherapy,* 1960, **10,** 675—681).—Root growth of young cucumber seedling and barley exposed to dil. solutions of hexachlorophene (**I**) was depressed by concn. as low as 2·7 μg./ml. while greater concn. inhibited growth. The mechanism of the injury is discussed. (11 references.) C. V.

Isolation and characterisation of malformin. N. Takahashi and R. W. Curtis (*Plant Physiol.,* 1961, **36,** 30—36).—Malformin, isolated from culture filtrates of *Aspergillus niger,* is a neutral peptide containing the amino-acids, valine, leucine, isoleucine and ½ cystine. The mol. formula $C_{23}H_{39}N_5S_2$ is proposed. E. G. BRICKELL.

Response of bean seedlings and maize roots to malformin. R. W. Curtis (*Plant Physiol.,* 1961, **36,** 37—43).—On bean stems malformin produced a visible response at 1×10^{-4} μg. per plant and optimum response at 1×10^{-1} μg. per plant. Malformin was translocated through the roots of the plants to the region of the apical meristem. With maize roots curvatures were obtained only when the tips were treated, optimum conditions being 1×10^{-3} μg. per root tip. Malformin was active on the above-ground portions of 20 out of 28 species of dicotyledons. On monocotyledons, only onion and Sudangrass responded out of the nine species tested. E. G. BRICKELL.

General effects of ethylene on enzyme systems in the cotton leaf. F. A. Herrero and W. C. Hall (*Physiol. Plant.,* 1960, **13,** 736—750).—Ethylene stimulated respiration and depressed the free sulphydryl content of both leaf blades and pulvini. It increased the activity of amylases, acid phosphatase, pectin esterase, peroxidase and catecholase but depressed catalase. Polygalacturonase, cellulase,

xylanase, IAA-oxidase, cellobiase and oxidative phosphorylation activities could not be demonstrated, possibly because of inactivation of these enzymes in the process of extraction. E. G. BRICKELL.

Biological activity of phenylboric acid. B. Haccius and D. Massfeller (*Planta*, 1961, **56**, 174—178).—Applications ($10 \times 0{\cdot}03$ ml.) of the acid (2×10^{-3}M) to the growing points of *Kalanchoe blossfeldiana* seedlings cause partial or complete inhibition of the development of petals. In *Cucumis sativus* one application suffices to inhibit the development of leaves as well as petals, and also to produce other abnormalities. (14 references.) P. S. ARUP.

Absorptiometric determination of fluoride in grass. Anon. (*U.K. Atomic Energy Authority*, 1960, PG Rep. 144[S], 8 pp.).—The grass is dried and ashed in presence of $Mg(OAc)_2$. A portion is then mixed with U_3O_8, pyrohydrolysed at 1000° and the F^- collected in NaOH. F^- is determined absorptiometrically by its bleaching action on Al Solochrome cyanine reagent. A. C.

Determination of phosphoric acid in plant material. R. Grossmann (*Ann. agron.*, 1960, **11A**, 357—360).—The ash of material rich in P and K and poor in Ca generally contains a large proportion of pyrophosphate, in which case special attention must be given to ensure the complete hydrolysis to orthophosphate. P. S. ARUP.

Crops and Cropping

Sowing dates, seed rates and nitrogen for spring wheat. N. Forbes (*Exp. Husbandry*, 1960, No. 5, 1—6).—In a 6-year study on a heavy calcareous clay soil, there was no evidence that a late sowing of Atle spring wheat required a heavier seed rate than usual. The yield of grain was not depressed by delaying drilling up to the end of April. Late sowing tended to reduce response to N, but allowed the seedbed to be cultivated for a longer period and effectively reduced the incidence of wild oats. E. M. J.

Effects of time of ploughing and time of drilling on development and yield of the winter wheat crop following and clover ley. F. Hanley, R. H. Jarvis and J. D. Whitear (*J. agric. Sci.*, 1961, **56**, 119—125).—The effects of different ploughing times arise from resulting differences in the physical conditions of the seedbed. Delay in drilling retards crop development but not yield irrespective of ploughing date. Low response to applied N suggests some other limiting factor in these trials. M. LONG.

Relation between pH, total silica and mobile silica in wheat grains. L. Delmas (*C. R. Acad. Sci., Paris*, 1960, **251**, 2402—2404).—Determinations are recorded of SiO_2 in ripe grain, and in extracts thereof (water, EtOH, $HClO_4$, trichloroacetic acid), from plots (i) permanently deprived of fertiliser, and (ii) treated with N, P, K or manure. Total SiO_2 (**I**) increased with decreasing pH of aq. extract, but mobile SiO_2 (**II**) generally increased with pH, its concn. per g. of dry wt. varying (490—540 μg.) with the nutrients. The ratio **II** : **I** decreased with pH. Plant metabolism probably ensures that **II** is a constant fraction of the absorbed SiO_2. From the same plot, those tissues having lowest pH have high **I** and generally low **II**. These findings explain the results of Whittenberger (*Amer. J. Bot.*, 1945, **32**, 539). W. J. BAKER.

Applying nitrogen fertilisers for spring barley. F. V. Widdowson, A. Penny and R. J. B. Williams (*J. agric. Sci.*, 1961, **56**, 39—45).—Combine drilling 0·3 and 0·6 cwt. of N/acre as $(NH_4)_2SO_4$ gives higher yields than broadcasting, but 0·9 cwt. of N/acre checks early growth so reducing yields. The lowest level supplied as a Nitrochalk top-dressing is superior to combine drilling or broadcasting but the middle level depresses yields. Above 0·6 cwt. of N/acre only small gains in yield are obtained and lodging is likely. Dressings above 0·3 cwt. per acre increase N content, lower dressings having little effect. The proportion of small grain in the sample is increased by N dressings. M. LONG.

Movement of chlorine in the rice kernel. III. Moisture in rice kernel as the dominant factor for the chlorine translocation. IV. Relations of the inner translocation of chlorine with increase of inorganic phosphorus and change in germination power. S. Kubo and K. Fujita (*Nippon Nôgei Kagaku Kaishi*, 1960, **34**, 572—576, 577—579).—III. Translocation of Cl^- in various samples of rice kernels occurred more quickly at high temp. and R.H. than at low temp. and high R.H. Movement of Cl^- did not occur at low R.H. Probably some free water is necessary for Cl^- translocation; the limiting water content for Cl^- to move into the endosperm was 14·5—15% at 22° or 15·5—16% at 5°. Translocation rates were influenced by variety of rice and locality of growth.

IV. The contents of inorg. P were determined on the above samples after 10 months' storage. The increase of inorg. P and decrease of germination power after storage at lower temp. (13—15°) appeared to run parallel, but they showed no correlation with the rate of Cl^- translocation. Samples stored at high temp. and lowest R.H., had a relatively high content of inorg. P and showed better germination than did those stored at lower temp. and high or low R.H. S. KAWAMURA.

Effects of irrigation, nitrogen level and plant population on maize yields. R. Vázquez (*J. Agric. Puerto Rico*, 1960, **44**, 121—127).—Irrigation did not affect maize yields when more than 20 in. of well-distributed rainfall fell during the season. Consumptive use of water increased to the tasseling–hard-dough stage and decreased thereafter. Yields were increased by the application of 80 lb., but were not increased further by 160 lb. of N per acre. Yields increased with plant population (9600—19,400 plants per acre) where irrigation was practised, but decreased where irrigation was not practised. There were significant interactions among irrigation, plant population, and level of N application. A. H. CORNFIELD.

Effects of fertilisers on the yield of potatoes in S.E. Scotland. K. Simpson and P. Crooks (*J. Sci. Fd Agric.*, 1961, **12**, 131—137).—The main effects of N, P and K on the ware, seed and total yield of potatoes (Majestic) grown with 10—15 tons/acre of farmyard manure were studied. Application of N (120 lb./acre) depressed yields of ware and seed; application of P (60 lb. of P_2O_5/acre) increased seed yield but had little effect in soils of high content of easily-sol P. KCl increased the ratio of ware/seed. Findings suggest lower rates of application of N, P and K than those at present used in Scotland for potatoes grown for seed in presence of farmyard manure. (14 references.) E. M. J.

Nitrogen supply of potatoes by means of farmyard manure and artificial fertilisers. IV. J. Kortleven (*Vers. landbouwk. Onderz.*, 1960, 65.19, 83 pp.).—The N of farmyard manure (FYM) applied in autumn acted, as regards yields, mostly as soil-N, whereas FYM applied in spring supplied (under unfavourable climatic conditions) only the N already present in the mineral form, behaving in this respect like Nitro-chalk. The initial effect of autumn FYM is reduced owing to leaching of the inorg. N during winter, but the mineral N derived therefrom during later growth is particularly effective as regards yields. (28 references.) P. S. ARUP.

Effect of soil compaction on potato growth. G. R. Blake, D. H. Boelter, E. P. Adams and J. K. Aase (*Amer. Potato J.*, 1960, **37**, 409—413).—Compacting a silty clay loam after seedbed prep. and before planting resulted in delayed emergence of potatoes and reduced plant vigour and yields, but had no effect on the number of hills at harvest. The treatment resulted in the tubers setting an average of 1 in. nearer the soil surface, but had no effect on the number of tubers set or on the % of mis-shapen tubers. Compaction resulted in the production of tubers of lower sp. gr. A. H. CORNFIELD.

Effect of foliar applications of chloride and sulphate on the specific gravity of white potato tubers. H. W. Gausman, G. O. Estes and R. A. Struchtemeyer (*Amer. Potato J.*, 1960, **37**, 377—378).—Applications of NaCl or Na_2SO_4 (Cl^- or SO_4^{2-} 150—600 p.p.m.; total Cl^- or SO_4^{2-} 0·98—3·93 lb. per acre) in three foliar sprays, commencing at bloom and at weekly intervals thereafter, were made to Katahdin potatoes. The Cl^- applications had no significant effect on sp. gr. of tubers at harvest. Applications of SO_4^{2-} 300—600 p.p.m. or of a combined SO_4^{2-}–Cl^- spray (300 p.p.m. each) significantly reduced tuber sp. gr. The amounts of Cl^- and SO_4^{2-} brought down by rain may have a significant effect on tuber sp. gr. A. H. CORNFIELD.

Greening and solanine development of white potato in fluorescent light. A. Liljemark and E. Widoff (*Amer. Potato J.*, 1960, **37**, 379—389).—When tubers were subjected to fluorescent light of different wave-lengths but of the same irradiation intensity chlorophyll development increased in the order green-, red-, blue-, daylight. Solanine development on the outer layers of the tubers gave inconsistent results, possibly due to movement of solanine to the inner parts of the tubers. A. H. CORNFIELD.

Effects of environment on tuber production, potassium absorption and susceptibility of potatoes to virus disease in Poland. A. Kozlowska (*Amer. Potato J.*, 1960, **37**, 366—372).—Yields of tubers decreased and tuber-K % (dry basis) increased with altitude of growth (up to 1300 m.). Seed tubers produced at high altitudes (>400 m.) gave high yields of tubers when planted at low altitudes and showed little spread of leaf roll and streak viruses. Seed pieces produced at altitudes <400 m. gave relatively low yields and had a high incidence of the virus diseases when planted at low altitudes. A. H. CORNFIELD.

Effects of viruses and other potato diseases on chip colour. H. W. Chapman and C. W. Frutchey (*Amer. Potato J.*, 1960, **37**, 257—259).—Tubers infected with leaf roll and spindle tuber viruses and tubers from plants showing vine symptoms of mosaic and calico viruses gave chips as light and uniform in colour as those from healthy controls. Tubers from giant-hill- and haywire-infected plants produced chips of a darker colour than those from healthy plants.

Blackleg-infected tubers produced chips with dark edges, whilst *Fusarium*-infection and the resulting discoloration produced a dark ring or portion of a ring in the chips. A. H. CORNFIELD.

Relationship of the chlorogenic acid and phenol oxidase content of a potato variety and its resistance to common scab. E. T. Holm and A. P. Adams (*Enzymologia*, 1960, **22**, 245—250).—The outer layers of seven varieties of potato, four resistant and three susceptible to scab, were assayed for chlorogenic acid and phenol oxidase content. No correlation was found between the contents of these substances and the resistance to scab. S. A. BROOKS.

Effects of antibiotic and fungicidal treatments on wound periderm formation, plant emergence and yields produced by cut seed potatoes. R. Bonde and F. Hyland (*Amer. Potato J.*, 1960, **37**, 279—288).—Treatment of freshly-cut seed potatoes with agrimycin (100 p.p.m., 5 min. dip) reduced the amount of bacterial seed-piece decay, but also reduced the rate and extent to which the wound protective layer was formed. Treatment with agrimycin + captan (2 lb. per 100 gal.) did not reduce the effect of the antibiotic on wound-periderm formation, and increased the susceptibility of the seed pieces to surface growth by moulds and to decay by *Fusarium* spp. The combined treatment had no effect on emergence or yields. A. H. CORNFIELD.

Effect of fertility level on the incidence of hollow heart of potatoes. A. Kallio (*Amer. Potato J.*, 1960, **37**, 338—343).—Incidence of hollow heart of tubers on a very fine sandy loam increased with level of applied N (60—180 lb./acre), decreased with increasing level of applied K_2O (120—360 lb.), and was not affected by level of applied P_2O_5 (210—630 lb.). A. H. CORNFIELD.

Sprouting, plant growth and tuber production as affected by chemical treatment of white potato seed pieces. II. Effect of temperature and time of treatment with gibberellic acid. H. Timm, L. Rappaport, P. Primer and O. E. Smith. **III. Compatibility of gibberellic acid with chemicals used for seed treatment.** H. Timm and L. Rappaport (*Amer. Potato J.*, 1960, **37**, 357—365, 403—408).—II. Treatment of potato seed pieces, from a crop harvested 2 weeks previously, with gibberellic acid (1—25 p.p.m., 5 min. dip) (**I**) prior to sowing resulted in increased rates of emergence of four varieties. The stimulating effect of **I** on emergence declined with length of time the treatment was given after harvest and was also low at low soil or storage temp. The treatments had no effect on plant growth or tuber characteristics.

III. Treatment of potato seed pieces with chemical disinfectants usually retarded emergence, whilst treatment with **I** (0·5—1·0 p.p.m., 5 min. dip) hastened it. The latter effect was not reduced when the disinfectant treatments were combined with **I**. Stem infection by *Rhizoctonia* was severe after treatment with **I**, but was absent after treatment with $HgCl_2$ (**II**). After initial whole-seed treatment with **II**, applying **I** to cut seed along with various fungicides tended to increase seed-piece decay, except where **II** was the fungicide. A. H. CORNFIELD.

Effect of time of application of phosphate and potash on sugar beet. S. N. Adams (*J. agric. Sci.*, 1961, **56**, 127—130).—P and K ploughed down in the previous autumn give less sugar and tops than when broadcast in spring, even in dry summers. Starter applications in the seedbed coupled with ploughing down of P and K yield almost as much as do spring applications. M. LONG.

Lime status of soil and yield of beets on sandy and sandy peat soils. C. M. J. Sluijsmans and K. Boskma (*Versl. landbouwk. Onderz.*, 1960, No. 65. 18, 39 pp.).—An investigation of results (representing 103 experimental years) in the possession of the Institute for Soil Fertility, Groningen. A mean curve for soil-pH against root-yield of mangolds and sugar beets, integrated from each experimental year, conforms (for pH >3·6) with the Mitscherlich equation. Immediate effects of liming are less beneficial than longer-term effects. Liming is more necessary and effective on poor than on rich soil. An inverse relationship is found between lime requirement and N-manuring, but not with K- or P-manuring. No correlation is found between the response of root-yield to increases in pH and the yield-level. (26 references.) P. S. ARUP.

Survey of productivity level. II. Fertilisation of grassland as practised in the Netherlands. J. Koopmans (*Versl. landbouwk. Onderz.*, 1960, No. 66.5, 112 pp.).—A statistical and critical review based on information received from ~1500 farms during 1950—2. (14 references.) P. S. ARUP.

Effects of fertilisers on herbage production. I. Effects of nitrogen, phosphate and potash on yield. J. W. S. Reith, R. H. E. Inkson, A. B. Stewart, W. Holmes, D. S. Maclusky, D. Reid, R. G. Heddle, D. Clouston and G. J. F. Copeman (*J. agric. Sci.*, 1961, **56**, 17—29).—The % dry matter in the herbage is reduced by heavy N dressings, whilst dry matter yields are increased, being nearly doubled by 348 lb. of N per acre. N response depends on an adequate K supply, very large interactions existing between the two. Heavy N dressings exhaust the soil-K reserves. P has practically no effect on dry matter yields, whether or not N was given. M. LONG.

Efficiency of recovery of applied nitrate by perennial rye-grass from different soils. A. R. Grable and D. D. Johnson (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 503—507).—The efficiency of utilisation of applied NO_3^- (N 200—600 lb./acre) by rye-grass from 14 soils was studied in pot tests. Yields of grass and total N uptake over 270 days (4 clippings) were proportional to the rate of applied N. The efficiency of utilisation of applied N averaged 77%. With some, but not all, efficiency of utilisation of applied N was slightly higher at high than at low levels of application. Efficiency was not related to the ability of the grass to take up native N, but was lowest on the heavier-textured soils. A. H. CORNFIELD.

Agronomic performance of a form of signal grass, *Brachiaria brizantha*, Stapf., compared with that of Guinea grass, *Panicum maximum*, Jack. A. Sotomayor-Rios, J. Vélez-Fortuño, R. Woodburg, K. F. Schertz and A. Sierra-Bracero (*J. Agric. Puerto Rico*, 1960, **44**, 208—220).—On a clay (pH 4·5) signal grass gave yields similar to those given by Guinea grass and also showed high drought resistance. Signal grass had slightly higher % of P and somewhat lower % of Ca, Mg and lignin than had Guinea grass. A. H. CORNFIELD.

Acetone-soluble lipids of grasses and other forage plants. I. Galactolipids of red clover (*Trifolium pratense*) leaves. R. O. Weenik. **II. General observations on the properties of the lipids with special reference to the yield of fatty acids.** F. B. Shorland (*J. Sci. Fd Agric.*, 1961, **12**, 34—38, 39—43).—I. The acetone-sol. lipids of red clover consist largely of galactolipids, galactosyl-1-glycerol and digalactosyl-1-glycerol linolenate. The fatty acids from the galactolipids as determined by gas–liquid chromatography consisted of linolenic 95·8, linoleic 1·9 and palmitic acid 2·3 moles-%, respectively. No free glycerol was detected in the neutral hydrolysates after saponification of the galactolipids with $Ba(OH)_2$. (23 references.)

II. The lipids sol. in acetone at 0° from the leaves of rye-grass (*Lolium perenne*), cocksfoot (*Dactylis glomerata*), white clover (*Trifolium repens*) and rape (*Brassica napus*, L.) were examined. The yields of fatty acids (corrected for unsaponifiable matter) were 70·5—76·5% as compared with 95·6% required for a pure triglyceride calculated as trilinolenin. When the cold acetone-sol. lipids of rye-grass were subjected to diffusion through a rubber membrane with light petroleum as solvent most of the fatty acids were present in the non-dialysable lipids (i.e., as galactolipids, as in red clover leaves). Galactolipids and not triglycerides were the main lipid constituents of the leaves examined. The dialysable lipids comprised 28·7% of the total acetone-sol. fraction; the non-dialysable lipids yielded linolenic acid in high concn., 88% of the total fatty acids. (18 references.) E. M. J.

Lime and soil acidity effects on lucerne growth in a Red-Yellow Podsolic soil. W. W. Moschler, G. D. Jones and G. W. Thomas (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 507—509).—Application of 1 ton of dolomite per acre, which increased soil pH from 4·9 to 5·7, greatly increased yields and survival of lucerne over 3 years. Heavier applications (up to 16 tons of dolomite or $CaCO_3$ per acre) had little further effects on yields. A considerable decrease in exchangeable Al occurred with small dressings of liming materials, and little further decrease with heavier dressings. Yields were better correlated (negatively) with soil exchangeable Al than with exchangeable Ca or Mg. Yields were also better correlated with % base saturation calculated from exchangeable(Ca + Mg + K)/exchangeable(Ca + Mg + K + Al) than from % base saturation calculated from exchangeable(Ca + Mg + K)/exchangeable(Ca + Mg + K + H). Depth of root penetration was not affected by liming, but extent of root growth in the surface soil was increased greatly. A. H. CORNFIELD.

Carotene stability in lucerne as affected by laboratory- and industrial-scale processing. C. R. Thompson, E. M. Bickoff, G. R. van Atta, G. O. Kohler, J. Guggolz and A. L. Livingston (*U.S. Dep. Agric. agric. Res. Serv.*, 1960, Tech. Bull. 1232, 14 pp.).—Apart from instability caused by pre-drying treatment in one case, all the samples of dehydrated forage examined were of comparable carotene stability. Blanching lucerne before drying had little effect on subsequent stability but continued oven heating after drying reduced both carotene content and stability. Ethoxyquin was superior to *NN*-diphenyl-*p*-phenylenediamine, 2,5-dibutylquinol, or butylated hydroxytoluene as an antioxidant. Increasing levels of animal tallow oil alone or with Ethoxyquin increased carotene retention. Stability did not appear to be linked with lucerne variety. (15 references.) E. G. BRICKELL.

Influence of cobalt on nitrogen fixation by *Medicago*. C. C. Delwiche, C. M. Johnson and H. M. Reisenauer (*Plant Physiol.*, 1961,

36, 73—78).—Co plays a major rôle in the lucerne (*Medicago sativa*)–*Rhizobium* symbiosis. With added Co and a *Rhizobium* inoculum significantly greater yields were obtained and roots and nodules possessed a greater capacity for N fixation. E. G. BRICKELL.

Reaction of lucerne seedlings to high concentrations of manganese. L. Dessureaux (*Plant & Soil*, 1960, **13,** 114—122).—The presence of Mn (1000 p.p.m.) did not reduce the germination of lucerne seedlings in vermiculite. [Mn] of 100 p.p.m. delayed leaf emergence, whilst 400 p.p.m. was toxic to seedlings. There were significant differences between lines of seed with respect to their ability to tolerate Mn, as indicated by size of the unifoliate leaf, dry matter yields and Mn content of the shoot. Maternal influences from reciprocal crosses were most important in assessing the reaction of seedlings to high Mn levels, but seed size was sometimes related to early growth characteristics. A. H. CORNFIELD.

Effect of molybdenum application on growth and composition of lucerne and distribution of molybdenum in a Cecil-Lloyd soil. J. Giddens and H. F. Perkins (*Proc. Soil Sci. Soc. Amer.*, 1960, **24,** 496—497).—Application of dolomitic limestone (500—4000 lb./acre) to a sandy loam in 1954 increased yields of lucerne in 1956—9 but not in 1955. Annual applications of Mo (Na molybdate 8 oz. per acre) to the limed plots increased lucerne yields, in comparison with those from unlimed plots, only in 1958—9. The Mo treatment increased N % and Mo % in the plants. There was little leaching of Mo into the subsoil. A. H. CORNFIELD.

Effect of pH and organic compounds on nitrogen fixation by red clover. E. G. Mulder and W. L. van Veen (*Plant & Soil*, 1960, **13,** 91—113).—Red clover grown on an acid soil was poorly nodulated and gave low yields due to N deficiency. Inoculation of the soil with an effective strain of *Rhizobium trifolii* greatly increased nodulation and dry matter yields of clover. Addition of $CaCO_3$ to non-inoculated soil brought about normal nodulation of clover, but only after about 4 weeks. There was no increase in effective *Rhizobium* no. in the $CaCO_3$-treated soil in the absence of red clover or in acid soil in the presence of red clover, but there was a great increase in $CaCO_3$-treated soil in the presence of red clover. Treatment of acid soil with yeast extract or a sterilised suspension of effective or ineffective *R. trifolii* increased nodulation of red clover. Stable manure and its extracts sometimes produced the same effect. A. H. CORNFIELD.

Dry matter and protein yields in four strains of *Leucaena glauca*, Benth. E. M. Hutton and I. A. Bonner (*J. Aust. Inst. agric. Sci.*, 1960, **26,** 276—277).—In trials near Brisbane, leaves and stems were harvested from trees in Nov., Jan., March and June. The dry matter and protein yields were calculated and compared favourably with yields from lucerne, and high quality clover–rye-grass pastures. S. G. AYERST.

Rest in apple trees.—W. H. Chandler (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 1—10).—The " rest " influence in apples in late summer, autumn and winter was sufficient to prevent growth even when the average mean temp. from Dec. 1st to Feb. 15th varied from 9·4 to 10·0° over 15 years, but by early spring it allowed brief impeded growth of very short shoots. Exposure to high temp. (6 h. at 44°—50°) was as effective as chilling (100 or more h. at 4·4°) in breaking the rest period. High temp. in late summer, however, increased chilling requirements. L. G. G. WARNE.

Effect of diurnal variation in sampling, and effect of methods of preparation and storage of samples on total nitrogen and mineral content of apple leaves. P. S. Lieu (*Dissert. Abstr.*, 1960, **21,** 720—721).—The nutrient element content of morning-picked leaves is a little, but not significantly, higher than that of afternoon-picked leaves. Samples should not be kept for >2—4 h. at room temp. before washing and drying. Tap-water and 0·025N-HNO_3 or -H_2SO_4 are recommended for washing. Significant differences between samples ground in the Wiley mill and those ground by porcelain mortar and pestle are noted for Fe and Cu, but not for the other elements. Samples dried by various methods give practically the same analytical results. Determinations of N should be made on the freshly-prepared samples. P. S. ARUP.

Effects of environment and chemical additives on absorption of dinitro-*o*-cresol by apple leaves. M. N. Westwood, L. P. Batjer and H. D. Billingsley (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 30—40).—The absorption of dinitro-*o*-cresol given as an aq. foliar spray to apples was increased by high R.H. before spraying but this effect was reduced if artificial rain followed the spraying. Absorption was increased when a suitable wetting agent was used and was greater at 27° than at 16° in the absence but not in the presence of a wetting agent. Absorption continued after the spray had dried. L. G. G. WARNE.

Effects of environment and chemical additives on absorption of naphthaleneacetic acid by apple leaves. M. N. Westwood and L. P. Batjer (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 16—29).—Absorption of naphthylacetic acid given as a foliar spray to apples was increased by high R.H. before or after spraying, by pretreatment with red light, by the use of soft water and by the addition of a wetter. The effects of the wetter were greatest under conditions when absorption would otherwise have been low. The addition of urea, bentonite and of insecticides to the spray prevented absorption. Sunlight had no effect on absorption which was greater on the under than on the upper side of the leaves and occurred mainly after the spray had dried. L. G. G. WARNE.

Effects of several organic spray materials on fruit growth and foliage efficiency of apple and pear. M. N. Westwood, L. P. Batjer and H. D. Billingsley (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 59—67).—Six but not three sprays of toxaphene or dieldrin reduced the growth rate of Bartlett pear fruits. " Black Leaf 40 " and malathion did not have this effect. Sprays did not affect apple growth when soil moisture was deficient but with ample moisture eight sprays of Aramite (but not of parathion, Kelthane or Tedion) reduced fruit growth and apparent photosynthetic rate. Sprays of Sulphenone or Aramite given to apple trees in the greenhouse reduced growth. Tedion, parathion and Kelthane had little effect on growth. L. G. G. WARNE.

Further comparisons of growth, maturity and quality of seedless and seeded Bartlett pears. W. H. Griggs, L. L. Claypool and B. T. Iwakiri (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 74—84).—Self pollinated (and generally seedless and hence parthenocarpic) Bartlett pears matured more slowly, and had lower sol. solids than seeded pears produced following cross pollination. L. G. G. WARNE.

Influence of gibberellin on resting pear buds. D. S. Brown, W. H. Griggs and B. T. Iwakiri (*Proc. Amer. Soc hort. Sci.*, 1960, **76,** 52—58).—Gibberellin sprays (250—2500 p.p.m.) were given to pear trees between Oct. 14th and Feb. 19th. In some trees subsequently brought into the greenhouse, spraying stimulated bud break only after the buds had received some winter chilling Especially with the early sprays many flowers within the clusters were killed and the fruit set was deformed at the calyx end. Trees which remained outdoors showed no response. L. G. G. WARNE.

Effects of principal nutrient elements on growth and fruiting of peach trees. J. Liwerant (*Ann. agron.*, 1960, **11A,** 93—111).—Annual applications of NPK result, after 14 years, in vigorous growth and increases in fruit yields by 102%. Omission of P reduces the increase in yields by 20%. The effects of PK or NP, alone, are much less satisfactory. (10 references.) P. S. ARUP.

Effects of salinity on four varieties of table grapes grown in sand culture. C. F. Ehlig (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 323—331).—Four grape varieties were grown in sand and supplied with mixed $CaCl_2$ + NaCl (1 or 2 atm. osmotic pressure, O.P.), $CaCl_2$ (2 atm.), NaCl (2 atm.), Na_2SO_4 (2 atm.) or no salt. High chloride caused leaf burn on all varieties and the injury developed much more rapidly at 38° than at 32°. $CaCl_2$ was more injurious than NaCl. Na_2SO_4 induced Mg deficiency. Varieties differed in their sensitivity to Cl^- and this variation reflected differences in the rate of Cl^- accumulation. L. G. G. WARNE.

Boron deficiency dieback in highbush blueberry. C. G. Woodbridge and R. H. Drew (*Plant Dis. Reptr*, 1960, **44,** 855—857).—A dieback of highbush blueberry was associated with low B % in the tissue. There were varietal differences in susceptibility to B deficiency. Foliar application of B sprays in the autumn prevented recurrence of the trouble the following two years and increased the B % in leaves and twigs. A. H. CORNFIELD.

Morphological aspects of growth in subtropical fruits. C. A. Schroeder (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 248—252).—Cell division continued in avocado pear fruits throughout the life of the fruit on the tree (in contrast to the tissue behaviour in fruits of apple, peach, cherry, plum and tomato). Mitotic divisions are most frequent between 11 p.m. and 1 a.m., corresponding with a period of rapid increase in fruit size. L. G. G. WARNE.

Influence of season and temperature on carbohydrates in avocado shoots. J. Rodrigues and G. F. Ryan (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 253—261).—Young shoots of avocado were high in sugars in winter in California. The concn. of sugar then decreased until autumn when it rose again. Young spring shoots were high in sugar in May. In detached shoots (at 1·7°) starch disappeared rapidly and the glucose concn. increased. This change also occurred at higher temp. (10°) but much more slowly. Whole young trees subjected to low temp. showed a decrease in starch concn. and an increase in sugar concn. L. G. G. WARNE.

Effects of *Phytophthora* spp. on water used by citrus seedlings. L. H. Stolzy, P. W. Moore, L. J. Klotz, T. A. DeWolfe and T. E. Szuszkiewicz (*Proc Amer. Soc. hort. Sci.*, 1960, **76,** 240—244).—Citrus seedlings in soil infected with *Phytophthora* spp. showed in the

second season a reduced water usage, presumably resulting from root destruction by the soil fungi. L. G. G. WARNE.

Effect of preharvest foliar sprays of maleic hydrazide on the cold storage behaviour of onions and garlic. W. B. Date (*Food Sci., Mysore*, 1960, **9**, 203—204).—Onions and garlic plants were sprayed with solution of maleic hydrazide in active ingredient 750, 1000 and 1500 p.p.m. 6 weeks before harvest. Samples were stored at 32-35°F and R.H. 80—90%. The higher dosages of maleic hydrazide showed little depressant action on respiration rate in onions or garlic. Onions showed 100% rooting within the first two months of storage, garlic showed none during storage for 180 days. The higher dosages of maleic hydrazide showed mild stimulant effects similar to those of the smaller dosages employed by Mathur *et al.* (J.S.F.A. Abstr., 1959, i, 24). I. DICKINSON.

Influence of fluorescent light quality on growth and photosynthesis of tomato. S. Dunn and F. W. Went (*Lloydia*, 1959, **22**, 302—324). —Of a great variety of commercially available lamps used to supply light to tomato plants, warm white fluorescent lamps augmented by incandescent lamps gave the highest growth rates and yields. Still better results were obtained with specially made blue and high-intensity red fluorescent lamps augmented by incandescent light. L. G. G. WARNE.

Manuring problems of forest soils. D. Lamberts (*Agricultura*, 1960, **8**, 231—246).—A review covering evidence of the low nutrient status of Belgian forest soils and experiments to determine suitable methods of manuring. (71 references.) P. S. ARUP.

Uptake of potassium by red pine seedlings and losses through leaching from fertilisers of various solubilities. H. H. Krause and S. A. Wilde (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 513—515).—Considerable amounts of K were lost from application of KCl to a coarse-textured soil. K losses over 5 months were correlated with NO_3^- losses. KPO_3 was less subject to leaching and had a greater fertilising value for pine seedlings than had KCl. Syenetic granite (20-mesh) was a satisfactory source of K only during the first season of growth. A. H. CORNFIELD.

Damping-off of black pine (*Pinus nigra*, Arn). D. S. Kailidis and F. C. Strong (*Mich. agric. Exp. Sta. Bull. quart. Bull.*, 1960, **43**, 14—32).—Germination of black pine at pH 4·5 was approx. double that at pH 8·5 and took place much more rapidly, thus lowering the incidence of damping-off. A. G. POLLARD.

Fertilisation of southern pines. W. L. Pritchett and W. K. Robertson (*Proc. Soil Sci. Soc. Amer.*, 1960, **24**, 510—512).—Applications to soil of ^{32}P-labelled PO_4^{3-} showed that 2-year- and 5-year-old pines fed to distances of 15 and 32 ft. respectively. Plots 0·25—0·5 acre are necessary to provide an adequate sample for measurements and borders to prevent cross-feeding. Application of sol. N or K_2O >300 lb./acre tended to reduce the growth and stands of newly-planted pines on a sandy soil. A. H. CORNFIELD.

Method for measuring the water consumption of larger intact trees. K. Ladefoged (*Physiol. Plant.*, 1960, **13**, 648—658).—A belt of the rising sap stream is heated 8—10° above the temp. of the surroundings by short-wave diathermy and temp. at standard intervals above the point of heating are followed by a sensitive thermo-element mounted outside the bark. From these the time t to reach max. deflection is correlated with the rate of the transpiration stream and then compared with a calibration curve relating t and water consumption (l./h.). This latter curve is initially prepared by sawing off a tree under water and measuring the uptake of water directly for different values of t. E. G. BRICKELL.

Determining critical ranges in leaf contents of nutrient elements from changes in gradients along axes of one-year-old tung trees. C. B. Shear, H. L. Barrows, M. S. Neff, B. G. Sitton and W. W. Kilby (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 310—322).—One-year-old tung trees were trained to various branching habits, and leaves collected from five positions on the trunk and three on lateral branches and analysed, and the gradients of nutrient concn. in the trunk and branch leaves examined. The thesis is advanced that "the critical range of an element normally supplied as a cation (K, Ca, Mg, Cu, Fe, Mn and Zn) is that range in leaf content at which the gradient of the element from basal to terminal leaves shifts from positive to negative or negative to positive." This does not hold for N, P or B when supplied as anions. L. G. G. WARNE.

Effects of time of applying fertilisers and levels of calcium and magnesium on growth and production of tung on Lakeland fine sand. M. S. Neff, H. L. Barrows and C. B. Shear (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 278—286).—In a Lakeland fine sand deficient in Ca and Mg, split applications of an NPK fertiliser, of dolomite and of Mg all increased growth. The split fertiliser treatment increased fruit no. and reduced the tendency to biennial bearing; the dolomite and Mg both increased leaf-Mg. All three treatments reduced leaf scorch. L. G. G. WARNE.

Effect of soil type and zinc concentration on growth, nutrient uptake and magnesium translocation of seedling tung trees. H. L. Barrows and N. Gammon, jun. (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 287—299).—Seedling tung trees were grown in a variety of soils to which differing applications of Zn had been made. Max. growth was correlated with leaf Zn equal to 0·41—0·77 mequiv. per 100 g. Top growth was more readily inhibited than root growth by excessive Zn. Concn. of Zn in leaf and root were positively correlated. The Zn affected Mg absorption and translocation but the effect depended on soil type. L. G. G. WARNE.

Response of one-year-old tung trees to levels and placements of zinc sulphate as affected by soil type. H. L. Barrows, M. S. Neff, N. Gammon, jun., and W. W. Kilby (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 300—309).—Zinc fertiliser trials on a variety of soil types are described. When $ZnSO_4$ was mixed with the soil in which the young trees were planted, tree growth and feeding root concn. in the soil were both reduced in three out of the four soils used. The amount of Zn needed to control Zn-deficiency varied with the soil. Zn concn. in trunk leaves was the most sensitive index of Zn uptake and this was highest when the $ZnSO_4$ was mixed with the soil rather than applied as a top dressing. L. G. G. WARNE.

Resistance of shoots of tung varieties to injury from controlled low temperatures. S. Merrill, jun., and G. F. Potter (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 270—277).—Susceptibility to low-temp. injury decreased throughout the autumn and increased throughout the spring. In late-blooming varieties the spring increase in susceptibility occurred later than in early-flowering types. L. G. G. WARNE.

Influence of fertilisers on cigar-filler tobacco quality. G. Samuels (*J. Agric. Puerto Rico*, 1960, **44**, 194—207).—Application of $(NH_4)_2SO_4$ (**I**) (N 50 lb./acre) to a clay increased tobacco quality. Rates up to 150 lb./acre had no further effect on quality. Application of lime (1000—2000 lb.) or MgO (300 lb./acre) increased quality over that affected by N applications alone. Other inorg. and org. sources of N were no more effective than was **I** in increasing quality. Leaf nicotine % increased with application of N up to 100 lb./acre. Application of P and K had no effect on quality but tended to reduce leaf nicotine %. A. H. CORNFIELD.

Effects of boron deficiency on cotton plant (*Gossypum hirsutum* L.). L. J. A. Neirinckx (*Ann. Physiol. veg., Brussels*, 1960, **5**, 1—18).—Experiments with sand cultures show the lack of B to have much more serious effects on growth and yields than lack of F, Mn, Zn or Cu. Among the symptoms described is the early appearance of "striated petiole." The mechanism of the physiological effect of B-deficiency is considered. (22 references.) P. S. ARUP.

Stimulation of yield [of rubber]: a comparison of proprietary yield stimulants. Anon. (*R.R.I. Plant Bull.*, 1960, 114—119).—Flomore and Ready Rub (containing 2,4,5-T) and Stimulex (containing 2,4-D) were studied in a large-scale experiment at Selangor. All three induced a satisfactory yield increase of approx. 30%. Stimulex proved to be less injurious to bark tissue than stimulants containing 2,4,5-T but induced damage to renewing bark when application was made above the cut. E. G. BRICKELL.

Growth and differentiation of *Atropa belladonna*, L. as affected by different sources of nitrogen. L. J. Schermeister, F. A. Crane and R. F. Voigt (*J. Amer. pharm. Ass., sci. Edn*, 1960, **49**, 694—697).—Plants, grown from seed to maturity in water culture, received varying amounts of NO_3^- and $NH_4^+ + NO_3^-$, with adequate amounts of other essential nutrients. Data indicated $NH_4^+ + NO_3^-$ administration to promote better growth. Plants fed with NH_4^+ had a greater dry wt. than those fed with NO_3^-. Shoot/root and leaf/stem ratios showed N usage to vary greatly at different levels of feeding. A. G. COOPER.

Nitrogenous constituents of *Atropa belladonna*, L. grown on different sources of externally supplied nitrogen. L. J. Schermeister, F. A. Crane and R. F. Voigt (*J. Amer. pharm. Ass., sci. Edn*, 1960, **49**, 698—705).—The N constituents in parts of plants receiving differing amounts of NH_3^- and $NH_4^+ + NO_3^-$ were examined. Free amino-acids formed <10% total N, the $NH_4^+ + NO_3^-$ source promoting formation of C_5 acids, and NO_3^- source that of C_4 acids. Protein formed <50% total N. The compositions of leaf and root protein were similar, but markedly different from that of the stem which served mainly as a reservoir of sol. N. Alkaloid concn. was greatest in roots receiving $NH_4^+ + NO_3^-$, coinciding with small amounts of free and combined proline (**I**). Large amounts of γ-aminobutyric acid (**II**) were also found. Possible rôles of **I** and **II** in alkaloid synthesis are discussed. (12 references.) A. G. COOPER.

Effect of soil temperature on the growth of *Phalaris tuberosa* L. H. J. Ketellapper (*Physiol. Plant.*, 1960, **13**, 641—647).—Top growth is strongly influenced by soil temp., the optimum being 20—25°. A constant temp. of 35° is very injurious to the plants and still higher temp. cause death. E. G. BRICKELL.

Pest Control

Use of radio-isotopes in biological investigation of fruit parasites. A. Soenen, M. de Proost and G. Vanwetswinkel (*Agricultura*, 1960, **8**, 259—268).—Out of 800—900 beetles (*Anthonomus pomorum* L.) tagged with ^{32}P by feeding, only 15 were located after 12 days. A migration of the beetles is indicated. (13 references.) P. S. ARUP.

Osmotic destruction of plant parasitic and saprophytic nematodes by the addition of sugars to soil. W. A. Feder (*Plant Dis. Reptr*, 1960, **44**, 883—885).—The addition of 5% of sucrose to soil samples gave complete or practically complete kill of nematodes after 24 h. Different species of nematodes varied in their susceptibility to the toxic action of sucrose and glucose. The extent of kill increased as the soil dried, probably due to the increasing osmotic effect of the soil solution. A. H. CORNFIELD.

Modification of the *Lemna* test for phytotoxicity. F. Fromm (*J. Agric. Puerto Rico*, 1960, **44**, 93—102).—A modification of the culture test using *Lemna minor* for assessing the toxicity of weed killers is described. A. H. CORNFIELD.

Metabolism of ^{14}C-labelled DDT in the larvae, pupae and adults of *Drosophila melanogaster*. D. B. Menzel, S. M. Smith, R. Miskus and W. M. Hoskins (*J. econ. Ent.*, 1961, **54**, 9—12).—Chromatograms from adults reared in a larval medium containing ^{14}C-DDT showed Kelthane to be greatest metabolite. There was also an unknown material similar to *pp'*-dichlorobenzophenone. This substance was absent from chromatograms of larvae and pupae. Topical application to adults showed that a DDT-tolerant strain produced large amounts of Kelthane as well as polar metabolites, while a DDT-susceptible strain produced no Kelthane. C. M. HARDWICK.

Effects of aldrin and DDT on soil fauna and arable land. C. A. Edwards and E. B. Dennis (*Nature, Lond.*, 1960, **188**, 767).—Considerable changes in population occurred in most groups of the soil fauna. Both insecticides significantly decreased the no. of Acarina. Aldrin also brought the population of Collembola to a low level, but DDT increased no. to a peak at 9 months after treatment, decreasing to a level slightly above that of the untreated plots. Dipterous and coleopterous larvae, pupae, thrips, pauropods and symphylids were all suppressed by both insecticides. Root aphids were greatly increased by aldrin but not by DDT. Neither insecticide affected small plant parasitic nematodes, earthworms or enchytraeid worms significantly. E. G. BRICKELL.

Physiological effect of carbamates on *Lepidium sativum*. F. Beye (*Flora*, 1960, **149**, 543—565).—Inhibitive activities (on a mol. basis) on germination, growth, respiration, catalase activity, and respiration rank in the (ascending) order: ethyl-, methyl- and phenylurethane, Isolan, Dimetan and Pyrolan. The possibility of specific effects, other than a general "narcotic effect," is considered, especially with regard to phenylurethane, the action of which appears to be more specific than that of the other insecticides. (32 references.) P. S. ARUP.

Bacterial wilt and Stewarts leaf blight of maize. A. L. Robert (*U.S. Dep. Agric.*, 1960, Fmrs' Bull., 1092, 13 pp.).—Distribution of the wilt disease caused by *Xanthomonas stewartii*, symptoms and control are discussed. E. G. BRICKELL.

Germination of small legume seeds after fumigation with methyl bromide and hydrocyanic acid. R. G. Strong and D. L. Lindgren (*J. econ. Ent.*, 1961, **54**, 21—25).—Seeds of *Medicago sativa*, *Trifolium hybridum*, *T. repens*, *T. pratense* and *Lotus corniculatus* at 6, 9 and 12% moisture content were fumigated for varying lengths of time, at different dosages and on one or two occasions. None lost their capacity for germination completely; only *M. sativa* was affected by HCN and only *L. corniculatus* was unaffected by MeBr. C. M. HARDWICK.

Preliminary assessment of toxaphene, Strobane and Thiodan for control of clover case-bearers (*Coleophora* spp.) (Coleophoridae, Lepidoptera). J. M. Hoy (*N.Z. J. agric. Res.*, 1960, **3**, 617—622).—Toxaphene and Strobane are highly toxic to adults only of *Coleophora* spp. but may be used while clover is in flower, with min. risk to honey bees. Thiodan (5 lb./acre) caused 60% larval mortality, but may be toxic to honey bees when used on flowering crops. E. M. J.

Disinfectants and potato ring rot control. D. S. MacLachlan (*Amer. Potato J.*, 1960, **37**, 325—337).—Of many materials tested for disinfecting the potato cutting knife 0·2% $HgCl_2$ and Semesan Bel (0·25 lb./gal.) were the most effective for controlling ring rot. Alkyltolylmethyl-trimethylammonium chloride (150 p.p.m.) was very effective for disinfecting jute bags. Ring rot incidence in the crop was sometimes high if cut seed pieces of a susceptible variety were stored in contaminated bags prior to sowing. A. H. CORNFIELD.

Late autumn applications of fumigant for the control of sugar beet nematodes. J. Altman and B. J. Fitzgerald (*Plant Dis. Reptr*, 1960, **44**, 868—871).—Application of D-D (1,2-dichloropropane + 1,3-dichloropropene, 14 gal./acre) in Nov. by injection into infested soil gave good control of nematode larvae the following season and doubled the yields of sugar beet. A. H. CORNFIELD.

Integrated control methods against Egyptian lucerne weevil in California. V. M. Stern (*J. econ. Ent.*, 1961, **54**, 50—55).—Heptachlor sprays (2—2·4 oz./acre) gave good control of larvae of *Hypera brunneipennis* and *H. postica* but did not affect their parasites; it was toxic to *Trioxys utilis*, a parasite of *Therioaphis maculata*, and *Chrysopa* spp. Demeton did not affect weevil larvae but was toxic to their parasites. C. M. HARDWICK.

Red clover mottle virus. R. C. Sinha (*Ann. appl. Biol.*, 1960, **48**, 742—748).—A virus, provisionally named red clover mottle virus, isolated from red clover plants, seemed distinct from any previously described. It was transmitted by mechanical inoculation of sap to many legumes and to *Gomphrena globosa* L., but was not transmitted by six aphid species or through soil or seeds. Some properties of the virus are presented. A. H. CORNFIELD.

Factors influencing citrus red mite populations on navel oranges and scheduling of acaricide applications in Southern California. L. R. Jeppson, J. O. Complin and M. J. Jesser (*J. econ. Ent.*, 1951, **54**, 55—60).—Populations of *Panonychus citri* build-up in spring, then decline in May and June and increase again in autumn and winter. Periods of high temp. and low R.H. delayed the autumn build-up. One spray application of Aramite or Kelthane in spring kept populations low if followed by one when the autumn build-up occurred. If the latter is early (Aug.) a third application may be necessary. C. M. HARDWICK.

Orchard-mite resistance to Kelthane. S. C. Hoyt and F. H. Harries (*J. econ. Ent.*, 1961, **54**, 12—16).—Sprays applied early in the year did not prevent the build-up of *Tetranychus medanieli* and later applications did not control them in various Washington orchards. Laboratory tests showed up to 200 × resistance. *T. telarius* from the same orchards did not show similar resistance. C. M. HARDWICK.

Rôle of attractants in recent Mediterranean fruit fly eradication programme in Florida. L. F. Steiner, G. G. Rohwer, E. L. Ayres and L. D. Christenson (*J. econ. Ent.*, 1961, **54**, 30—35).—The use of angelica seed oil or Siglure + DDVP in traps placed at 10—40 per sq. mile caught only 1 in 25 *Ceratitis capitata*. Aerial spraying of 5% protein hydrolysate + 25% malathion at 1 gal./acre depressed populations in 6—9 days. Up to nine applications were needed for eradication. Some damage to paint and some poisoning of fish in small ponds were reported. Populations of house flies and mosquitoes were also reduced. (20 references.) C. M. HARDWICK.

Effects of some insecticides on nitrogen fractions of Bartlett pear leaf tissue. A. L. Kamal and C. G. Woodbridge (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 68—73).—DDT sprays increased total- and protein-N in Bartlett pear leaves. Malathion, parathion and Systox had a similar but smaller effect as had oil and lime-sulphur when applied together (but not when applied separately). L. G. G. WARNE.

Control of bacterial spot of peach with n-dodecylguanidine acetate. R. H. Daines (*Plant Dis. Reptr*, 1960, **44**, 826—827).—Of a no. of materials tested against bacterial spot of peach, due to *Xanthomonas pruni*, the most effective control was given by dodecylguanidine acetate (**I**) (1 lb. per 100 gal.). Application of captan (1 lb./acre) in addition to **I** did not further improve control. A. H. CORNFIELD.

Heat treatment to destroy fungi in infected citrus seeds. L. J. Klotz, T. A. DeWolfe, C. N. Roistacher, E. M. Nauer and J. B. Carpenter (*Plant Dis. Reptr*, 1960, **44**, 858—861).—A 10-min. dip of citrus seeds in water at 51·6° eliminated *Phytophthora citrophthora* and *P. parasitica* without impairing germination. A. H. CORNFIELD.

Stubborn and other diseases of citrus in Morocco in 1959. J. F. L. Childs and J. B. Carpenter (*Plant Dis. Reptr*, 1960, **44**, 920—927).—Symptoms of stubborn disease were observed in all citrus regions. The disease may be transmitted by insects. The extent of the disease was not related to poor soil structure, bad drainage or other soil conditions, but the symptoms were accentuated by damaging the roots or by unbalanced fertilisation. Suitable fertilisation and controlled cropping reduced the symptoms. Psorosis and foot-rot were also common. Zn-deficiency (frenching) symptoms were common, particularly where N-fertilisers were applied. Lime-induced chlorosis and Mg- and Mn-deficiences were also observed. A. H. CORNFIELD.

Methods of application of emulsifiable 1,2-dibromo-3-chloropropane around citrus trees. S. D. Van Gundy, F. J. Foote, R. L.

Rackham and A. Rinkov (*Plant Dis. Reptr*, 1960, **44**, 830—833).—Basin irrigation for the application of 1,2-dibromo-3-chloropropane in citrus orchards produced the most uniform distribution of the fumigant and control of the citrus nematode. Overhead sprinkler distribution gave poor dosages under the trees and furrow irrigation was also unsatisfactory. A. H. CORNFIELD.

Gloeosporium musarum, Cke. & Massee causing storage rots of Jamaican bananas. III. Control with sodium salicylanilide (Shirlan WS) and nystatin. D. S. Meredith (*Ann. appl. Biol.*, 1960, **48**, 824—836).—Dipping inoculated fruit in 0·5—1·5% Shirlan WS for 1 min. gave 30—65% control of finger-stalk rot, but was highly phytotoxic. Rinsing with water following dipping gave 15—40% control without phytotoxic effects. Relatively poor control was obtained when the treatment was delayed for more than 24 h. following inoculation. Treatment with nystatin (200—400 p.p.m.) did not control finger-stalk rot. A. H. CORNFIELD.

Control of diseases of shallots. R. Aycock and J. M. Jenkins, jun. (*Plant Dis. Reptr*, 1960, **44**, 934—939).—Treating *Botrytis*-infested shallot bulbs with Dowicide B (Na 2,4,5-trichlorophenoxide, 1·75—3·00 lb.) or captan (12 lb. per 100 gal.) prior to planting increased yields by 50%. Soil treatment with pentachloronitrobenzene (50 lb./acre) also increased yields by 50% due to control of *Sclerotium rolfsii*. Drying bulbs in the field prior to storage resulted in less storage losses than did artificial drying. Treatment of bulbs with Dowicide B prior to storage reduced the incidence of black mould, *Aspergillus niger*. A. H. CORNFIELD.

Fumigation of agricultural products. XVII. Control of _Ascochyta_ blight of peas by fumigation. J. Kennedy (*J. Sci. Fd Agric.*, 1961, **12**, 96—103).—Fumigation with chloropicrin of diseased seeds with a moisture content of 14—16% for 72 h. at 20° was most satisfactory. In quartered seeds with moisture content of 14·2% fumigation for 24 h. gave almost complete fungal kill, and for 9 h. gave results similar to those for whole seeds fumigated for 96 h., showing the slowness of penetration of the fumigant. Infected seed was more severely damaged by fumigation than was healthy seed. The vigour of plants grown from treated seeds was normal. E. M. J.

Inhibition of the multiplication of tobacco mosaic virus in tissue culture by its antiserum. K. A. Miczynski (*Ann. appl. Biol.*, 1960, **48**, 739—741).—Extracts from tobacco tissue cultures infected with tobacco mosaic virus grown in medium containing anti-serum to the virus had only about 6—50% as much virus as extracts from tissues grown in media without the anti-serum. When tissues grown with anti-serum were washed thoroughly before they were extracted, the extracts contained as much virus as did extracts of tissues grown without anti-serum. The anti-serum did not affect virus multiplication, but antibodies in the tissues may have precipitated virus either in the cells or when the tissues were macerated. A. H. CORNFIELD.

***Fusarium* wilt of cotton. I. Factors influencing incidence.** J. Meyer (*Agricultura*, 1960, **8**, 203—218).—Published data are reviewed. An account is given of the operative factors observed by the author in the Congo, viz., exceptional abundance of *Fusarium* spores in the soil, infection by the agency of nematodes, and increased susceptibility due to K-deficiency. (38 references.) P. S. ARUP.

Nemic parasites of coffee in Guatemala and suggested _ad interim_ control measures. B. G. Chitwood, and C. A. Berger (*Plant Dis. Reptr*, 1960, **44**, 841—847).—Five genera and nine species of nemic parasites are thought to be potential pathogens of coffee. *Ad interim* control measures are suggested. A. H. CORNFIELD.

Cotton leaf perforator and its control in Arizona. D. M. Tuttle, G. P. Wene and L. W. Sheets (*J. econ. Ent.*, 1961, **54**, 67—70).—The 18-day life cycle of *Bucculatrix thurberiella* is described and its predators are listed. In general, sprays were more effective than dusts. Org. P compounds and chlorinated terpenes were more effective when used with DDT than when applied alone. C. M. HARDWICK.

Sorghum diseases and their control.—R. W. Leukel, J. H. Martin and C. L. Lefebvre (*U.S. Dep. Agric.*, 1960, Fmrs' Bull., No. 1959, 46 pp.).—The symptoms of the more important diseases are described together with practicable control measures by seed treatment. E. G. BRICKELL.

Control of tadpole shrimp, _Triops longicaudatus_ in California rice fields. A. A. Grigarick, W. H. Lange and D. C. Finfrock (*J. econ. Ent.*, 1961, **54**, 36—40).—In laboratory and field tests aq. $CuSO_4$ was highly toxic to the shrimp. Poor control was usually due to type of crystals used or to application practices. DDT (granules or spray) at 2 lb./acre gave satisfactory control. Diazinon and malathion had less residual effects. Sevin and Kepone were promising. C. M. HARDWICK.

Fluctuations in aquatic insect populations associated with aerial applications of DDT to Northern Maine forests. J. G. Gorham (*Dissert. Abstr.*, 1960, **21**, 716—717).—Sprayings for control of spruce budworm in June caused temporary reductions of ~50—65% in insect populations of streams in the sprayed areas, but had no long-term effects. Populations of most of the trout-food groups were completely or partly restored within two months. P. S. ARUP.

Effect of variations in formulation or application procedure on control of imported fire ant with granular heptachlor. C. S. Lofgren, V. E. Adler and W. F. Barthel (*J. econ. Ent.*, 1961, **54**, 45—47).—The no. of insecticide granules used did not affect the efficiency of heptachlor against *Solenopsis saevissima v. richteri*. Control was slower with coarser mesh granules and with calcined as against regular granules. Two applications with $\frac{1}{4}$—$\frac{1}{2}$ lb. of heptachlor or one with $\frac{1}{8}$—$\frac{1}{4}$ lb./acre gave nearly 100% control for 1 year while an application at $\frac{1}{8}$ lb./acre gave 79% control. C. M. HARDWICK.

Field studies with baits against _Solenopsis saevissima v. richteri_, the imported fire ant. F. J. Bartlett and C. S. Lofgren (*J. econ. Ent.*, 1961, **54**, 70—73).—The acceptance of non-toxic baits was determined by the stains on paper from ants squashed after feeding on dyed baits. Edible oils were very attractive but dried vegetable meals were not. Toxic baits had the insecticide sprayed on them in methylene chloride solution. Kepone gave a 92% reduction in colonies after 12 weeks. Re-infestation makes further treatment necessary. C. M. HARDWICK.

Control of spider mites and black spot on roses with acaricide–fungicide sprays and dusts. T. J. Henneberry, E. A. Taylor, F. F. Smith, A. L. Boswell and W. D. McClellan (*J. econ. Ent.*, 1961, **54**, 61—63).—Roses treated with malathion dusts and demeton sprays alone or with ferbam had fewer *Tetranychus telarius* than had untreated checks. Better control of *Diplocarpon rosae* was given by the addition of maneb and zineb rather than ferbam or captan to the dusts or sprays. Spider mite control resulted in increased yields in Sept. C. M. HARDWICK.

Control of spadix rot of anthurium, _Anthurium andraeanum_, in Hawaii. M. Aragaki and M. Ishii (*Plant Dis. Reptr*, 1960, **44**, 865—867).—*Colletotrichum gloeosporioides* was the causal organism of anthurium spadix rot, a previously unreported disease. The disease, principally affecting the rudimentary perianth, developed rapidly under favourable conditions of high temp. and humidity. Maneb (2 lb.), Dodine (1 lb.) and Dyrene (1 lb. per 100 gal.) as protective fungicides and hot-water treatment at 46—48° for 15 or 30 min. gave excellent control of the disease. A. H. CORNFIELD.

Pathological alterations in flax through inappropriate use of herbicides. E. Patitz (*Faserforsch. u. Textiltech.*, 1960, **11**, 495—500).—Discussions are given of the proper concn. and time of spraying of salts of 2-methyl-4-chlorophenoxyacetic acid for herbicidal use in the cultivation of flax. Use of unsuitable prep. or excessive concn. causes poor fibre yield, low tearing strength of the fibre and abnormally high lignin content. H. L. WHITEHEAD.

Chemical weed control with chloro-aminotriazine. G. Ubrizsy and M. Csongrády (*Acta agron. Acad. Sci. hung.*, 1960, **10**, 197—227).—Weeds in maize may be controlled by pre-emergence application of Simazine [2-chloro-4,6-bis(ethylamino)]- or Atrazine (2-chloro-4-alkylamino-6-isopropylamino-)-*s*-triazine, at the rate of 9—10 kg. per hectare. Two crops of maize in succession may be grown if weeds are thus controlled. A. G. POLLARD.

Microbial decomposition of halogenated propionic and acetic acids. P. Hirsch and M. Alexander (*Canad. J. Microbiol.*, 1960, **6**, 241—249).—Soil micro-organisms decomposing 2,2-dichloropropionic acid and related herbicides included *Nocardia* sp. and *Pseudomonas* sp. Liberation of Cl^- was substantially completed in 3 weeks. In other cases decomposition without formation of Cl^- probably occurs. Rates of decomposition were higher for α- than for β-substituted propionates. A. G. POLLARD.

A soil _Achromobacter_ which degrades 2,4-dichlorophenoxyacetic acid. G. R. Bell (*Canad. J. Microbiol.*, 1960, **6**, 325—337).—Oxidation of 2,4-D analogues and 2,4-dichlorophenol in soil was effected by *Achromobacter* spp. Structural formations associated with such oxidation includes, a free (especially unchlorinated) *o*-position, a free carboxyl on the side-chain (preferably β- with respect to the ethereal linkage), a Cl atom in the *p*-position and ≯2 atoms of Cl in the ring whether the *o*-position is free or not. A. G. POLLARD.

Determination of 2,6-dichloro-4-nitroaniline in Ditranil and fungicidal formulations by infra-red spectroscopy. P. G. Marshall (*Analyst*, 1960, **85**, 681—683).—For the samples, the residue from evaporation of an ethereal extract is dissolved in $CHCl_3$, the i.r.

spectrum is recorded at 1145 cm.$^{-1}$ and compared with that of a standard solution of 2,6-dichloro-4-nitroaniline. A. O. JONES.

N-monoaryl substituted carbamic acid 2,4,5-trichlorophenyl esters. E. Merck A.-G. (B.P. 834,358, 2.12.57. Ger., 4.12.56).—Compounds, useful as fungicides comprise 2,4,5-trichlorophenyl esters of R·NH·CO_2H (R is Ph optionally substituted by Me, Cl or NO_2). 2,4,5-*Trichlorophenyl phenylcarbamate*, m.p. 126—128°, is prepared by a conventional process. It (50) may be ground with kaolin (40) and a crumbly mixture (10 pt.) of C_{8-20}-fatty acids and poly-ethyleneglycolsorbite-oleate, to provide a spray-powder for use as a fungistatic. The action of the ester towards *Fungi imperfecti* is four times that of hypochlorite solutions. F. R. BASFORD.

Phosphorous acid esters. Farbenfabriken Bayer A.-G. (B.P. 834,392, 4.7.58. Ger., 10.7.52).—Compounds, useful as insecticides (highly active against flies, mites, aphids and caterpillars and red spiders), comprise derivatives OR·PY·S·X (R is alkyl of 1—4 C; X is alkyl radical substituted by aliphatic- or aromatic-thioether groups; Y is OR or radical of a secondary amine). Twenty-three compounds are specifically claimed. The prep. of OO-*diethyl* S.2.*ethylthioethyl phosphorothioite*, b.p. 40—45°/0·1 mm., is detailed A 0·001% solution is 100% lethal to red spiders and their eggs. F. R. BASFORD.

Thiophosphoric acid esters. Farbenfabriken Bayer A.-G. (B.P. 834,331, 26.7.57. Ger., 28.7.56 and 27.2.57).—Compounds $(OR)_2P(:X)\cdot X\cdot CH(SR')\cdot Cl_3$ and insecticidal compositions containing them are claimed (R is aliphatic radical of 1—4 C; X is O or S, but at least one X is S; R′ is alkyl, aryl or cycloalkyl). As an example of prep., condensation product of chloral and EtSH is treated with $(OEt)_2POCl$ and then S in toluene to give *Et*$_2$ 2,2,2-*trichloro*-1-*ethylthioethyl phosphorothionate*. F. R. BASFORD.

Thiophosphoric acid esters. Farbenfabriken Bayer A.-G. (B.P. 834,117, 6.12.57. Ger., 10.12.56).—Compounds claimed comprise thiophosphoric acid esters $(OR)_2\cdot P(:X)\cdot Y\cdot CH(CH_2\cdot A\cdot R')\cdot CH_2\cdot A\cdot R''$, useful as insecticides (R is alkyl of 1—4 C; R′ and R″ are alkyl or aryl; X and Y are O or S, at least one of these being S; A is SO or SO_2). As an example, OO-*diethyl* S-1,3-*di*(*ethylsulphinyl*)*prop*-2-*yl phosphorothioate* (prep. described) is 100% lethal to black bean aphids when applied as 0·1% solution. F. R. BASFORD.

Oxazoline and oxazine derivatives, polymers and copolymers thereof. Rohm & Haas Co. (B.P. 834,308, 7.5.57. U.S., 11.5.56).—A large range of oxazoline and oxazine deriv. is made useful *inter alia*, as fungicides against, e.g., *Macrosporium sarcinaeforme* and *Sclerotinia fructicola*. Details are given for the prep. of 2-*isopropenyl*-4,4-*dimethyloxazoline*, b.p. 58—59°/24 mm. H. S. R.

Phytotoxic and insecticidal triazines and preparations containing them. N.V. Philips' Gloeilampenfabrieken (B.P. 835,850, 2.4.58. Neth., 5,4,57).—Compounds, useful as active ingredients in phytotoxic and insecticidal conditions, comprise 4,6-dichloro-*s*-triazines substituted in the 2-position by $X\cdot R\cdot Y_n$ (X is O or S; R is alkylene of 2—7 C; Y is OR′, SR′, CO_2R′ or Cl; R′ is alkyl of 1—7 C; *n* is 1—2). 4,6-*Dichloro*-2-(3′-*chloropropoxy*)-s-*triazine*, b.p. 114—116°/0·07 mm., is prepared; it is 100% lethal to tomato and bush bean plants when applied thereto as a 1% solution in acetone at the rate of 10 kg. per hectare. F. R. BASFORD.

Chlorobenzoic amides and herbicidal compositions containing them. Hooker Chemical Corp. (B.P. 834,880, 8.2.56. U.S., 29.4.55. Divided out of B.P. 833,218).—A herbicidal composition, useful in the pre-emergent or post-emergent treatment of vegetation, comprises a carrier (preferably liquid, e.g., oil or water) and an amide of 2,3,5,6-tetrachlorobenzoic acid, prepared by interaction of the acid chloride with aq. NH_3. F. R. BASFORD.

Control of plant pests. Farbwerke Hoechst A.-G. (B.P. 835,546, 10.1.56. Ger., 10.1.55).—Compounds SnRR′R″R‴ are claimed as pesticides of high activity (at least one of R′—R‴ is diethyldithiocarbamato, ethylenebis-dithiocarbamato, pentachlorophenoxy, 2,4,6-trichlorophenoxyaceto, mercaptobenzthiazolo, [trichloromethyl-mercapto-dicarbethoxy]methyl, or *p*-toluenesulphonamido, the other radicals being alkyl). A typical prep., which is 100% effective against algae, comprises an aq. emulsion containing 0·5% of tributyl-(2,4,6-trichlorophenoxy)stannane. F. R. BASFORD.

Derivatives of 2-mercaptopyridine-1-oxide. Olin Mathieson Chemical Corp. (Inventors: J. Bernstein and K. A. Losee) (B.P. 834,553, 24.2.56).—Compounds, R_2S, R_2SO, R_2SO_2, R_2S_2, $RSO\cdot SR_2(RSO)_2$ and $RSO_2\cdot OR$ are claimed (R is a pyrid-2-yl-1-oxide radical optionally substituted by 1 or 2 halogen or low-mol. alkyl or alkoxy groups). They are made by treating an NH_4, alkali or alkaline earth metal salt of 2-mercaptopyridine-1-oxide with a 2-halogenopyridine-1-oxide and then oxidising with H_2O_2, or oxidation of a disulphide with H_2O_2. The compounds have antifungal properties and are useful in combating plant diseases (e.g., *Perenospora* on grapevines. 2,2′-*Dithiopyridine*-1-*oxide* has m.p. 200—201° (decomp.). H. S. R.

Animal Husbandry

Nutrition of zebu cattle. I. Equipment for the separate collection of urine and faeces from steers. R. M. Bredon, B. Marshall and C. D. Juko. **II. Techniques of digestibility trials with special reference to sampling, preservation and drying of faeces.** C. D. Juko, R. M. Bredon and B. Marshall. **III. Digestibility techniques: effects of combination of dry faeces, length of trials and number of animals required.** R. M. Bredon, C. D. Juko and B. Marshall (*J. agric. Sci.*, 1961, **56**, 91—92, 93—97, 99—103).—I. Urine is collected in a canvas funnel, held up by two straps, and faeces in a wooden box.

II. A 300-g. sample made up of 10-g. sub-samples is satisfactory. No significant difference in dry matter nor in crude protein contents is detectable between samples preserved with acetic acid, HCl or toluene, and dried at 80°, and those untreated and dried at 105° for 8 h. or those left at air temp. for 7 h. prior to drying. Where *X* and *Y* are the respective percentages of crude protein in dry and wet faeces, $Y = 1{\cdot}1252X + 0{\cdot}12$.

III. After a 14-day acclimatisation, a composite sample over a period of 10/12 days is obtained by mixing sub-samples the wt. of which is in proportion to the total dry matter of the individual collections. Foodstuffs were weighed and sampled each day for dry matter content, giving daily dry matter intakes, but composite samples for chemical composition are sufficient. A max. number of six steers only could be accommodated in the trials. M. LONG.

Comparative efficiency of digestion of feeds by ruminants and pigs. J. Glover (*J. agric. Sci.*, 1961, **56**, 113—115).—Relationships between % crude protein and crude fibre contents of feed dry matter and the different digestibilities of crude protein and total nutrients by both classes of animals lead to a classification of feeding stuffs which is in agreement with that based on practical experience. M. LONG.

***In vitro* production, by rumen micro-organisms of volatile fatty acids from cellulose and hemicullulose labelled with ^{14}C.** I. H. Bath and M. J. Head (*J. agric. Sci.*, 1961, **56**, 131—136).—Grass grown in $^{14}CO_2$ and fermented in an artificial rumen, shows that hemicellulose and α-cellulose both give rise to acetic and propionic acids in the molar ratio ~2 : 1. Small quantities of butyric, valeric and caproic acids are also found. M. LONG.

Estimation of the average total digestible nutrients in pig feeds. J. Glover and H. W. Dougall (*J. agric. Sci.*, 1961, **56**, 117).—Estimated values showing the very marked effect of increasing crude fibre in depressing the digestibility of total nutrients in pig feeds are given. M. LONG.

Development of a method for routine testing of poultry feeds. J. G. Miller and J. Edmondson (*N.Z. J. agric. Res.*, 1960, **3**, 633—640).—The trials reported have led to a suitable test for chick feeds and are being used in tests on feeds for adult birds, laying mashes, etc. A logical sequence of small trials, each designed on information obtained in the preceding one, economically provides the estimates of means and variances required in setting up a suitable test procedure which will have its probability of error below a given level. E. M. J.

Routine analysis of carbohydrates and lignin in herbage. R. E. Deriaz (*J. Sci. Fd Agric.*, 1961, **12**, 152—160).—The dried, wilted samples were extracted with diethyl ether (72 h.); sol. carbohydrates (**I**) were extracted with hot 0·5% NH_4 oxalate (2 h.) and structural carbohydrates (**II**) with N-H_2SO_4 followed by 72% H_2SO_4. **I** were determined with anthrone and the pentosans and hexosans derived from **II** were determined with aniline acetate and chromotropic acid, respectively; errors involved are discussed in detail. A value for lignin was obtained by determining loss in wt. on ignition of the residue and a correction applied for N × 6·25 after Kjeldahl analysis of the residue of another sample of the same herbage. (29 references.) E. M. J.

Influence of preservatives on fermentation, nutrient recovery and feeding value of lucerne, Starr millet, and cowpea and Sudan grass silages. M. E. McCullough, L. R. Sisk, O. E. Sell, A. R. Stasch and D. L. Cason (*J. Dairy Sci.*, 1960, **43**, 1826—1932).—The efficiencies of preservatives in preventing losses (and promotion of lactic acid fermentation) during the ensiling of first-cut lucerne are in the (descending) order: ground snap-maize (with 7% loss), citrus pulp, molasses (both with ~16% loss) and Na metabisulphite. The qualities are satisfactory except where Na metabisulphite is used.

No special difficulties are encountered with the other crops. In feeding trials with silages showing variations in the type of fermentation or losses, the differences are largely masked by grain supplementation. P. S. ARUP.

Feeding trials with sweet Sudan grass at the State Farms in the Comitat Tolna (Fornád, Alsópél) in 1958. J. Bajai (*Acta agron. Acad. Sci. hung.*, 1960, **10**, 1—40).—The high food value of the grass as a forage is emphasised and its economic use is discussed. No practical evidence of toxic effects of HCN derived from the grass were observed. A. G. POLLARD.

Digestibility studies on Venezuela grass, *Paspalum fasciculatum*, and plantain pseudostalks, *Musa paradisiaca*. J. A. Arroyo and L. R. Brenes (*J. Agric. Puerto Rico*, 1960, **44**, 103—106).—Plantain pseudostalks were lower in dry matter, crude protein and gross energy than was Venezuela grass, but the former were more palatable, based on feed consumption and dry matter intake. Protein digestibility of Venezuela grass was higher, whilst energy digestibility was lower, than that of plantain pseudostalks. A. H. CORNFIELD.

Availability and metabolism of various substrates in ruminants. III. Effect of propionate on acetate oxidation *in vivo*. C. L. Davis, R. E. Brown and J. R. Staubus (*J. Dairy Sci.*, 1960, **43**, 1783—1787; cf. *ibid.*, 241).—Addition of Na propionate to NaOAc (1-^{14}C) injected into the jugular vein of a steer reduces the total amount of CO_2 exhaled by 23·5%, but does not alter the ratio of $^{14}CO_2$ (derived from the acetate) to the total CO_2. P. S. ARUP.

Relation between protein nutrition and feed formulation. H. W. Bruins and R. O. Nesheim (*Cereal Sci.*, 1960, **5**, 312—316).—The amino-acid requirements of monogastric animals are considered in relation to feed formulation. The amino-acid content of grain, vegetable proteins, animal products and synthetic products and the reaction between these products during manufacture, are discussed. (12 references.) S. G. AYERST.

Residues in tissues and eggs of poultry dusted with Co-Ral (Bayer 21/199). H. W. Dorough, U. E. Brady, jun., J. A. Timmerman, jun., and B. W. Arthur (*J. econ. Ent.*, 1961, **54**, 25—30).—Hens were dusted once or twice with Co-Ral (50 mg./kg.). Half the Co-Ral disappeared from the feathers within 3 days of one application and 7 days of two applications. More ^{32}P was found in the bone than in the liver or fat. Acetonitrile-sol. residues were found in gizzard, liver and kidney but only in small quantities 3 days after one application and 7 days after a second one. In eggs the highest concn. of ^{32}P residues was in the yolk after 6—9 days. Acetonitrile-sol. derivatives were extremely low in the yolk and absent from rest of the egg. (13 references.) C. M. HARDWICK.

New anthelmintic for canine hookworm. R. W. Burrows, P. Clapham, D. A. Rawes, F. C. Copp and O. D. Standen (*Nature, Lond.*, 1960, **188**, 945—946).—The quaternary ammonium compound, *NN*-dimethyl-*N*(2-thienylmethyl)-*N*-(β-phenoxyethyl)-ammonium *p*-chlorobenzenesulphonate, was highly effective against both natural and experimental infections of *Ancylostoma caninum* and *Unicinaria stenocephala* in the dog, and natural infections of *Toxocara canis* and *Toxascaris leonina*. It combines high efficiency with only mild emetic properties. S. A. BROOKS.

Alimentary yeasts. Fromageries Bel-La Vache Qui Rit (B.P 835,544, 20.6.56. Fr., 22.6.55).—A process for the prep. of a burst alimentary yeast from buttermilk and/or whey (which has been deproteinised by pptn. of fermentable N-containing products, e.g., proteins, especially lactalbumin, by adding lactic acid at 80° to pH 4·5—4·7, then heating during 5 min. at 92—94°, decanting, and filtering) comprises subjecting the deproteinised filtrate to fermentation with the aid of a biological association of two or three yeast stocks selected from *Saccharomyces lactis Bel, S. lactis Dombrowski*, and *S. fragilis Jörgensen* or of two stocks of *Saccharomyces* and a stock of an imperfect form of *Saccharomyces*, viz., *S. lactis Dombrowski, S. fragilis Jörgensen*, or *Candida pseudotropicalis* (Cast.) Basgal var. *lactosa* (Harrison), Diddens et Lodder (to absorb and transform the carbohydrates), then simultaneously drying and bursting the (separated) yeast. The product is of use as in B.P. 835,545 (following abstr.) F. R. BASFORD.

Compound foods for calves. Fromageries Bel-La Vache Qui Rit (B.P. 835,545, 21.6.56. Fr., 23.6.55 .—A compound food for calves comprises an alimentary mixture (73) of powdered whey (46), milk protein powder (18—22), yeasts grown in lactic medium (see preceding abstr). (5—9 kg.), hydrolysed wheat fraction containing the wheat germ (1 pt. per 3 pt. of alimentary mixture), and mineral substances. F. R. BASFORD.

2.—FOODS

Carbohydrate Materials

Cereals, flours, starches, baking

Biochemistry of grain storage. P. Linko (*Cereal Sci.*, 1960, **5**, 302—306).—A review of recent work at Kansas University, on gas exchange, enzyme activation, free and bound water, browning and germ damage, and glutamic acid decarboxylase activity as a quality index of wheat. (26 references.) S. G. AYERST.

Microflora of cereals under various conditions of purification and grinding. G. Spicher (*Getreide u. Mehl*, 1960, **10**, 125—128).—The effects of storage for varying times and at varying temp. on the bacterial count and mould content of wheats are studied and portrayed in three-dimensional diagrams. J. V. RUSSO.

Chemical differentiation of proteins of wheat and rye. IV. Enzymic breakdown of wheat flour proteins and the resulting peptides. M. Rohrlich and W. B. T. Schulz (*Z. Lebensmitt-Untersuch.*, 1960, **113**, 361—369).—Column-chromatographic separation of the peptic hydrolysates of gliadin, glutenin, " haft-protein " and " zwickel-protein " resulted in characteristic curves of generally similar shape, although the proteins differed in their proportions of acid and of neutral and basic peptides. Examination of the products of acid hydrolysis of single or combined peptide fractions showed that peptides with up to 32 amino-acid residues can appear on enzymic breakdown of wheat proteins. The amino-acid composition of a characteristic peptide peak occurring regularly in wheat protein hydrolysates suggests at least a partial similarity of the different components of wheat gluten. (27 references.) C. L. HINTON.

Effect of bisulphite and acetaldehyde on the disulphide linkage in wheat protein. H. Matsumoto, I. Oshima and I. Hlynka (*Cereal Chem.*, 1960, **37**, 710—720).—Wheat proteins extracted from flour were treated with bisulphite. The increase in sulphydryl content was followed titrimetrically and the effects on it of pH level, acetaldehyde, oxidising agents and specific sulphydryl reagents were observed. Farinograms show, after reaction of the dough with bisulphite, that addition of acetaldehyde at pH 5 had little effect, but at pH 7 consistency was increased. (20 references.) S. G. AYERST.

Distribution of the amino-acids of wheat in commercial mill products. F. N. Hepburn, W. K. Calhoun and W. B. Bradley (*Cereal Chem.*, 1960, **37**, 749—755).—The 18 commonly occurring amino-acids were determined by microbiological analysis in two blends of wheat and in each of the 11 final products obtained by commercial milling. The distribution of the amino-acids in these fractions was examined. The amino-acid distribution of flour proteins was independent of the degree of separation of the flour as shown by the uniformity of amino-acid concn. within the flour fractions. The consistency of amino-acid distribution in bread flour protein may be related to the selection and blending of wheats. S. G. AYERST.

Distribution of the vitamins of wheat in commercial mill products. W. K. Calhoun, F. N. Hepburn and W. B. Bradley (*Cereal Chem.*, 1960, **37**, 755—761).—Nine vitamins were determined in a commercial wheat blend and in the eight products milled from it commercially. The pattern of distribution of the vitamins was investigated and data are given. (15 references.) S. G. AYERST.

New results on the chemical and enzymic degradation of phytin. J. Schormüller (*Getreide u. Mehl*, 1960, **10**, 137—140).—The prep. and ion-exchange separation of the inositol phosphoric acid esters are described in detail. The rate of enzymic degradation of each of these esters is studied. The monophosphate is found to degrade most slowly, probably due to steric hindrance. (14 references.) J. V. RUSSO.

Mono and oligo-saccharides in ripening grain. M. Rohrlich and W. Essner (*Getreide u. Mehl*, 1960, **10**, 121—125).—Fructose, glucose, maltose, sucrose and glucofructosan are determined paper chromatographically in rye and wheat at various stages of maturity of the grain, and the results are discussed. (15 references.) J. V. RUSSO.

Enzymic conversion of barley carbohydrate into syrup. M. J. Houle and K. J. Goering (*Food Technol.*, 1961, **15**, 25—28).—The conversion of whole barley into syrup by amylolytic enzymes is discussed. The composition of the syrup may be varied by selection of enzymes and conversion time. Barley contains substantial amounts of β-amylase so that only small additions (0·5%) of enzyme from a culture of *Aspergillus oryzae* were needed. Indications are that a high maltose syrup comparing favourably with maize syrup may be obtained. (11 references.) E. M. J.

Grain size determination in flours. K. Leschonski (*Getreide u.*

Mehl, 1960, **10**, 140—144).—Sieving and sedimentation methods for the determination of grain size in flours are compared and discussed. J. V. Russo.

Trypsin inhibitor in wheat flour. E. M. Learmonth and J. C. Wood (*Chem. & Ind.*, 1960, 1569—1570).—Commercial flours had 0·4—0·6% of the anti-trypsin activity of enzyme-active soya. The trypsin inhibitor of wheat is probably associated with the protein fraction of the flour. The activity persists during 3 h. fermentation but is almost completely destroyed by baking. A. C.

Suitable processing methods for spoiled rye flour from germinated grain. A. Schulz and H. Stephan (*Brot u. Gebäck*, 1960, **14**, 240—245).—Processes for obviating the effects of spoiled rye flour from germinated grain in bread production are described in detail. Rye flours type 997 and 812, of low enzyme activity, are recommended. The addition of 20% wheat flour or 5—10% starch has an improving effect. Long baking times at lower temp. are also advisable. (11 references.) J. V. Russo.

Measurement of the improver response in dough. I. Hlynka and R. R. Matsuo (*Cereal Chem.*, 1960, **37**, 721—727).—Structural relaxation data were obtained for doughs having four levels of bromate and three reaction times. When the semi-axis constant for bromate effect was plotted against reaction time, linear plots were obtained. Analogous plots were obtained when the amount of bromate reacted in dough (determined chemically) was plotted against reaction time. The amount of change in the semi-axis constant per unit amount of bromate reacted was taken as a definition of bromate response. The response was evaluated for several flours. Improver response was also obtained from similar data with iodate. S. G. Ayerst.

Action of amylases in baking. H. D. Ocker (*Brot u. Gebäck*, 1960, **14**, 245—250).—The influence of specially prepared mould and bacterial amylases on the baking quality of two flours of vastly different natural enzyme activity and of otherwise similar characteristics is studied. (25 references.) J. V. Russo.

Aromatic compounds present in oven gases. L. Wiseblatt (*Cereal Chem.*, 1960, **37**, 728—733).—Compounds identified in bread oven vapour condensates from loaves were ethanol, acetaldehyde and acetic acid. Mixtures of these needed furfural and acetylmethylcarbinol added in order to approach the natural aroma of oven gases. An "optimum" composition is given. Other compounds, at present unidentified, are present in the natural oven vapours. S. G. Ayerst.

Volatile organic acids found in dough, oven gases, and bread. L. Wiseblatt (*Cereal Chem.*, 1960, **37**, 734—739).—The concn. of volatile aliphatic acids (acetic, n-butyric, isovaleric and n-caproic) found in fermented dough, bread, and oven gases were estimated by vapour-phase chromatography. The amount of acids in bread, and oven gases, does not equal that in the dough. The fate of the dough acids during baking is considered. S. G. Ayerst.

Sulphydryl losses during mixing of doughs: comparison of flours having various mixing characteristics. H. A. Sokol, D. K. Mecham and J. W. Pence (*Cereal Chem.*, 1960, **37**, 739—748).—Twelve varied types of flour were made into doughs and the sulphydryl contents determined after mixing for 2, 5, 10 and 20 min. Considerable differences in the rate of sulphydryl loss were found. An initial short period (2—5 min.) of rapid decrease in sulphydryl content was observed with most flours. The effect of added flour albumin on the rate of loss and the correlation of the loss with mixing curve characteristics were examined. When doughs were mixed with added flour albumin the initial decrease in sulphydryl content was more rapid. (10 references.) S. G. Ayerst.

Analysis of food flavours by gas–liquid chromatography. Separation and identification of neutral components from bread pre-ferment liquid. D. E. Smith and J. R. Coffman (*Analyt. Chem.*, 1960, **32**, 1733—1737).—Twenty-seven ether-sol. neutral components in pre-ferment brew are separated and isolated by gas chromatography and identified by i.r. spectroscopy, mass spectrometry and deriv. formation. Initial tests are made for ethyl formate, γ-butyroacetone, 1,3-propanediol monoacetate and β-phenylethanol. (16 references.) C. B. Baines.

Problems in sliced bread packing. F. Zeppelzauer (*Brot u. Gebäck*, 1960, **14**, 250—252).—The optimum conditions and packing materials for the production of packed sliced bread of good keeping quality are discussed. The use of Al foil as a packing material is recommended. J. V. Russo.

Flour quality requirements for the preparation of fancy pastries. A. Rotsch and E. Tessmer (*Brot u. Gebäck*, 1960, **14**, 233—236).—Strong, protein-rich flours produce the best and lightest fancy pastries. J. V. Russo.

Effect of flour-fraction interchange on cake quality. D. H. Donelson and J. T. Wilson (*Cereal Chem.*, 1960, **37**, 683—710).—The water-solubles, gluten, starch tailings and prime starch, of a good and a poor cake flour, were combined to form several reconstituted flours. The effects of this interchanging on the vol. and structure of cakes baked from these flours were examined. Prime starch from poor flour was significantly superior to that of good flour. Gluten had the greatest effect on cake vol. and structure. Interactions of gluten × composition and water-solubles × tailings × composition were highly significant, which indicated considerable variation of responses to these flour components with concn. S. G. Ayerst.

Keeping quality of biscuits fortified with proteins and vitamins. D. S. Bhatia, N. S. Kapur and K. M. Narayanan (*Food Sci., Mysore*, 1960, **9**, 280—281).—"Nutro" biscuits were stored for 7 months in paper and in tin containers at room temp. and at 37°. Samples were withdrawn every month and the moisture, carbonyl value, thiamine, vitamin A, pH and free fatty acids were determined. All biscuits were acceptable organoleptically after 7 months; storage in tins was better than in paper. There was no appreciable loss in thiamine, riboflavin and nicotinic acid. Vitamin A decreased progressively, the loss in biscuits stored in paper at room temp. was 34·2%, that at 37° was 47·3%. In the biscuits in tins the loss of vitamin A was 28·9% and 44·7% respectively. I. Dickinson.

Conditioning of grain. A.-B. Svenska Fläktfabriken (B.P. 834,738, 27.3.58. Swed., 29.3.57).—In the conditioning of granular material, e.g., grain (prior to storage), by subjecting it in the form of a fluidised layer on a supporting plane to a process of drying and then a process of cooling by means of a gaseous medium (forced upwards through the material layer), the material between the two stages is caused to pass through an equalising zone in which it is substantially isolated from the influence of the gaseous medium and is thereby rendered uniform prior to subsequent treatment. Apparatus is figured. F. R. Basford.

Sugars and confectionery

Ion-exchange method for determining cations and the ash content of sugar products. S. Zagrodzki and H. Zaorska (*Roczn. Technol. Chem. Żywności*, 1960, **5**, 5—15).—The sum of K, Na, Ca and Mg ions of the sugar product is taken as the basis of sugar ash calculations. The sample solution is passed through an ion-exchange column filled with a polystyrene resin containing active SO_3H groups and treated with aq. NH_3. The K and Na ions are extracted with $(NH_4)_2CO_3$, and are later determined by titrating the extract with HCl. The Ca and Mg ions are extracted with 25% aq. NH_3 portionwise, and are determined by titrating a known amount of EDTA (disodium salt) solution with the second extract. The Fe or Al ions, if present, are individually determined in the second extract. The new method takes only 90 min., and after the second extraction the ion-exchange column is ready for the next determination. The so-called carbonate or sulphate ash content can be calculated from the titrations. (11 references.) A. L. Grochowski.

Surface-active agents: Their function and application in confections. W. H. Knightly (*Mfg Confect.*, 1961, **41**, No. 2, 29—34).—A general review relating to the glycerol mono-esters, lecithin, sorbitol, mannitol, etc. The rate of hardening in starch-based confectionery is retarded, loss of gloss and bloom formation is decreased while sticking in caramel and nougat is reduced. Aerated products are more readily whipped, and oil migration, especially in groundnut products, is diminished. In certain cases, e.g., mints, palatability is increased. (11 references.) C. V.

Purification of cane sugar solutions. Inventa A.-G. für Forschung u. Patentverwertung (B.P. 834,988, 18.8.58. Switz., 28.10.57).—A neutral crude sugar solution is treated with a solid water-insol. metal oxide (e.g., α-Fe_2O_3, MnO_2) at 50—100°, to absorb coloured impurities. F. R. Basford.

Amyloglucosidase. A. E. Staley Mfg Co. (B.P. 834,334, 6.8.57).—Amyloglucosidase, useful in the production of crystal dextrose from starch, is obtained by subjecting aq. nutrient medium in presence of *Aspergillus phoenicis* ATCC 13,156 or 13,157 to submerged aeration. F. R. Basford.

Galactomannan gum solutions. Stein, Hall & Co. Inc. (B.P. 834,375, 31.3.58. U.S., 1.4.57).—The degradation of a galactomannan material (e.g., guar gum) in the presence of water at elevated temp., and at pH >5·5, is retarded by the addition of a salt from a water-sol. metal sulphite, thiosulphate or arsenite (e.g., zinc sulphite) to a colloidal aq. solution of the material. E. Enos Jones.

Fermentation and Alcoholic Beverages

[A] **Factors influencing uptake of copper from brandy by ion-exchange resins.** [B] **Lead content of Australian brandies.** B. C. Rankine (*J. Sci. Fd Agric.*, 1961, **12**, 188—194, 194—196).—[A] The cation but not the anion resin efficiently removed the Cu in young brandy in the H and Na form of the resin. In old brandy efficient removal of the Cu occurred only with the cation resin in the H form. Very little Cu was removed from Cu complexes with caramel and tannin at pH >5. Quality was slightly impaired by ion-exchange treatment. Plastic tubing produced a permanent haze. In a survey of Australian brandies (33 representative samples) the Cu content was 0·7—12 p.p.m., mean value 3·4 p.p.m.

[B] The Pb contents of Australian brandies (37) ranged, with one exception (0·25 p.p.m.), from <0·01 to 0·06 with a mean of 0·029 p.p.m. These included samples from high-strength bulk brandy shipped to the United Kingdom and subsequently bottled. The Pb content of Australian brandies is therefore well within the proposed British legal limit of 0·5 p.p.m. E. M. J.

Sulphurous acid, aldehyde and biological reduction of acid in large-volume [must] fermentation and storage. F. Prillinger (*Mitt. Weinu. Obstbau, Wien*, 1960, **10 A**, 130—137).—The only difference between the results of fermentation in large tanks and in casks lies in the better clearing effects (due to mechanical factors) observed in the casks. The behaviour and effects of SO_2 and acetaldehyde are explained. Premature oxidation of a large proportion of the SO_2 (depending on the oxidising capacity of the must and the time elapsing before the onset of fermentation) can cause undesirable results, including excessive biological consumption of the acids. The importance of correct timing of the addition of metabisulphite is stressed. Sulphurisation to 50 mg. per l. of SO_2 is generally necessary and not excessive. P. S. Arup.

Influence of sulphurous acid and L-ascorbic acid in wine-making. II. Inactivation of polyphenoloxidases. W. Diemair, J. Koch and D. Hess (*Z. LebensmittUntersuch.*, 1960, **113**, 381—387).—The polyphenoloxidases (**I**) of grape juice can be inactivated by addition of SO_2 (50—100 mg./l., before fermentation) as well as by flash heating. The inactivation is reversible and time-dependent, and is not linked with any oxidation of the SO_2. It can be suppressed by other reactions such as combination of the SO_2 with CH_3CHO; and addition of CH_3CHO in excess can partly restore the **I** activity. In grape juice this activity is associated with cloudiness. It diminishes slowly during storage of the juice, and during fermentation disappears in 3 or 4 weeks. The sulphydryl compounds formed during fermentation either block the active prosthetic Cu of the **I** or precipitate it as sulphide. (13 references.) C. L. Hinton.

Utilisation of ascorbic acid in the conservation of wines. R. Cordonnier (*C. R. Acad. Agric. Fr.*, 1960, **46**, 745—747).—The effects of the addition of ascorbic acid to prevent " casse oxydasique " and oxidation of white wines are studied and compared with the use of SO_2 as an antiseptic. Ascorbic acid alone is useless as a " casse oxydasique " preventor, but is very effective when mixed with SO_2. The addition of ascorbic acid (40—60 mg./l.) to wines previously stabilised with K sorbate prevents oxidative deterioration (after storage for 6 months) more effectively than SO_2 addition. J. V. Russo.

Examination of sparkling wines. I. Objective tests for quality. A. Janke and M. Röhr (*Mitt. Wein- u. Obstbau, Wien*, 1960, **10 A**, 111—123).—A method is described whereby the vol. of CO_2 given off during 10 min. by the wine on raising the temp. from 1° to 20° is determined as a % of the total vol. given off at 20° and 35° (K^{20}_{10}). The test sample is taken by means of a 10-ml. pipette from the bottle after stabilisation at −2° and slowly releasing the pressure; the sample (2 × 10 ml.) is stabilised at 1° before making the test. Out of a total of 53 commercial samples, the bottle-fermented wines give fairly consistent values of $K^{20}_{10} > 30$, whilst tank-fermented wines give less consistent higher values. Values for Kjeldahl-N determined on solids obtained by membrane-filtration (representing autolysed yeast) are, for the former, ~55% > those obtained for the latter wines. The value of autolysed yeast as a quality-factor is considered. (10 references.) P. S. Arup.

Colorimetric determination of iron in wine by sulphosalicylic acid. H. Konrad (*Nahrung*, 1960, **4**, 365—373).—Current methods are critically examined. The method of Eberius (cf. *Angew. Chem.*, 1951, **63**, 513) (described) compares favourably with the German official iodometric method as regards rapidity, reproducibility, and the size of the sample required. The non-interference of the ions present in the ash of wine is confirmed. (19 references.) P. S. Arup.

Production of beer by continuous fermentation. J. S. Hough (*Soc. chem. Ind.*, 1961, Monogr. 12, 219—229).—Recent work is reviewed. (21 references.) A. J. B.

Mono-, oligo- and polysaccharides of pressed- and brewers' yeast. K. Täufel, K. J. Steinbach and G. Meinert (*Nahrung*, 1960, **4**, 295—309).—Current methods are discussed and applied to the extraction, paper-chromatographic separation, and determination of the saccharides. Results (%) obtained for pressed- and brewers' yeast are, respectively: trehalose 12·2, trace; mannan 12·4, 10·8; glycogen 20·2, 18·8; glucan 6·6, 9·1; xylan 1·9, 2·3; an unidentified trisaccharide nil, 11·4. The trisaccharide is non-reducing, and probably based on glucose (2 mol.) and fructose (1 mol.). (12 references.) P. S. Arup.

Quantitative determination of flavouring oil content of hop cones. H. Bausch, E. Rothenbach and H. Wolter (*Nahrung*, 1960, **4**, 599—618).—Factors affecting the yield were examined and a routine analytical method was developed. Details are given of a steam distillation method (of finely powdered hop cones, 5 g.), in which the condensate is passed through a 10-cm. layer of pentane, 98% of the yield being absorbed. The residue is examined turbidimetrically. By steam distillation of selectively extracted hops, substances other than flavouring oil (**I**) influencing the yield were obtained. To obtain hops free from **I** the hexane-sol. resins and **I** were extracted with hexane. The quantity of water to be distilled is fixed. The method is reproducible, sensitive to ±0·02% and is completed in 2·5 h. (32 references.) E. M. J.

Corrosion in breweries. R. H. Chandler (*Corrosion Prev. Control*, 1961, **8**, No. 1, 32—34).—The use of resistant materials, cathodic protection and protective coatings are briefly reviewed. C. V.

Continuous fermentation process. J. Labatt Ltd. (B.P. 835,964, 16.10.57. U.S., 4.3.57).—A process for the continuous fermentation of a yeast-fermentable substrate to produce especially a potable fermented beverage (from brewer's wort) comprises maintaining separated yeast-propagation and product-forming stages; maintaining aerobic conditions, favourable to yeast propagation in the yeast propagation stage (which is operated at 45—90°F); maintaining anaerobic conditions favourable to EtOH formation in the product-formation stage (which is preferably operated at 45—90°F, but 10°F below the temp. of the yeast propagation stage); continuously introducing fermentable substrate to the yeast-propagation stage; continuously passing effluent therefrom to the product formation stage; separating yeast from effluent of the product formation stage; and reintroducing thereto a controlled amount of the separated yeast to maintain a controlled concn. (equivalent to 5—15% of pressed cake). F. R. Basford.

Fruits, Vegetables, etc.

Quality and condition changes of McIntosh apples stored in controlled atmospheres. G. D. Blanpied and D. H. Dewey (*Mich. agric. Exp. Sta. quart. Bull.*, 1960, **42**, 771—778).—Storage of the apples in a controlled atm. (CO_2 5, O_2 3%) at 38°F was more effective than that in air at 32—33°F. Differences in fruit quality (firmness, texture, flavour, ripening) under the two storage conditions were not apparent until after 100 days but increased subsequently. The rate of decline in quality after removal from storage was similar in both cases. The incidence of core-browning, storage scale and mealy breakdown was much less in fruit stored in the controlled atm. A. G. Pollard.

Control of superficial scald on Granny Smith apples using pre-harvest sprays of diphenylamine. C. A. S. Padfield and R. M. Smock (*N.Z. J. agric. Res.*, 1960, **3**, 675—679).—Pre-harvest spraying of trees with diphenylamine (**I**) (500—1500 p.p.m. in aq. suspension) reduced subsequent development of scald on apples in cool store, but post-harvest treatment with **I** gave better results especially for New Zealand fruit intended for long storage. E. M. J.

Rôle of apple cuticle in development of storage scald on Cortland apples. V. G. Shutak and E. P. Christopher (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 106—111).—Puncturing the cuticle of Cortland apples decreased the development of scald in store. L. G. G. Warne.

Acceptance of apples by a taste panel as related to chemical composition. N. Harris and A. L. Kenworthy (*Mich. agric. Exp. Sta. quart. Bull.*, 1960, **43**, 149—153).—Apples stored for a month at 32°F were transferred to temp. of 75°F for periods up to 6 days. Taste-panel data indicate relationships between sensory tests and the total and acid-sol. pectin contents and (inversely) the total reducing-sugar contents. Sensory tests for texture were probably directly related to respiration rate, water-sol. pectin and titratable acidity and, inversely, to non-reducing sugar and sugar/acid ratios. Flavour and aroma were related to total pectin, acid-sol. pectin and, inversely, to reducing and total sugar contents. No relation between taste and chemical factors was apparent. A. G. Pollard.

Factors affecting quality of canned apple sauce. R. C. Wiley and V. Toldby (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 112—123).—Apple sauce quality is affected by variety, length and type of storage before processing and maturity. Quality tests in the raw apples did not indicate their value for sauce manufacture. L. G. G. WARNE.

Fresh fruit temperatures and transit injury. N. F. Sommer, F. G. Mitchell, R. Guillou and D. A. Luvisi (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 156—162).—Vibration increased respiration and wt. loss and invasion by decay fungi in pears. These effects were greater at high than at low temp., but dropping damaged cold fruits more than warm ones. L. G. G. WARNE.

Shrinkage of cherries in jelly. M. J. Anthistle (*J. Sci. Fd Agric.*, 1961, **12,** 208—211).—Cherries set in a gel containing gelatin, sugar and citric acid, sterilised by heat treatment, shrivel after storage for a few weeks. This is most likely caused by difference in osmotic pressure on either side of the cherry skin and an osmotic flow of liquid from the cherry into the gel. The shrinkage depends on the isoelectric pH of the gel and becomes less as that pH is approached ; it is discussed in terms of Donnan's theory of membrane equilibrium. E. M. J.

Use of light transmittance techniques to estimate the chlorophyll content and stage of maturation of Elberta peaches. A. P. Sitwell, G. S. Birth, J. V. Ernest and C. Golumbic (*Food Technol.*, 1961, **15,** 75—78).—Measurements of peaches from successive harvests with three different light transmittance instruments are reported and compared with the chlorophyll content of the fruit. Frozen slices of peaches which had been classified with one of the instruments were rated for eating. (15 references.) E. M. J.

Moisture determination in dates by forced ventilation infra-red drying. G. Zimmermann (*J. Sci. Fd Agric.*, 1961, **12,** 240—246).—Methods of determining moisture in dates for quality control, that are simple, reliable and give reproducible results in reasonably short time are discussed. I.r. drying with forced ventilation was applied to five varieties of dates ; 5-g. samples and a 250-W lamp were used. Heating was for 10 min. at 50 W and then 15 min. at 60 W with a vac. of 160 mm. of Hg and a distance between lamp and sample of 30 mm. Results compared with those obtained by standard vac. drying and by toluene distillation (reference method) were highly reliable and reproducible. (20 references.) E. M. J.

Effects of temperature, washing and waxing on composition of the internal atmosphere of orange fruits. I. L. Eaks and W. A. Ludi (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 220—228).—The CO_2 concn. of the internal atm. of orange fruits is greater in fruit held at 20° than at 10° or 0° whilst the O_2 concn. is lower. Washing and waxing the fruit also increased the CO_2 and decreased the O_2 concn. but had little effect on the respiration rate at 20°. L. G. G. WARNE.

Precooling, packaging and fungicides as factors affecting appearance and keeping quality of oranges in simulated transit experiments. W. Grierson and F. W. Hayward (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 229—239).—Precooling of orange fruits often decreased initial losses due to decay in transit but increased losses during the subsequent " consumption period." A low transit temp. had a persistent beneficial effect. Fungicidal treatment achieved a greater reduction in decay than did refrigeration unless this was continued right up to the final sale. L. G. G. WARNE.

Changes in the physico-chemical composition of mangoes during ripening after picking. G. V. Krishnamurthy, N. L. Jain and B. S. Bhatia (*Food Sci., Mysore*, 1960, **9,** 277—279).—Hard green mangoes of four different varieties were picked, stored and analysed at four different stages of ripening. Brix at 20°, water-insol. solids, total solids, pH, acidity, β-carotene μg., glucose, fructose, sucrose and total sugars were determined. The colour and the flavour of the flakes were assessed. Total solids of the pulps at all stages of maturity remained constant. Sol. solids, pH, β-carotene and sugar increased while acidity and water-insol. solids decreased rapidly with progressive ripening. Good quality flakes could be obtained from fully ripened mangoes only. (12 references.) I. DICKINSON.

Effect of acid level, calcium salts, monosodium glutamate and sugar on canned pimentos. J. J. Powers, D. E. Pratt, D. L. Downing and I. T. Powers (*Food Technol.*, 1961, **15,** 67—74).—The addition of citric acid (**I**) or various Ca salts (**II**) significantly increased drained wt. and firmness of the canned product ; when **I** and **II** were added, the effect was synergistic. Monosodium glutamate (0·05%) added to acid-treated pimentos, increased organoleptic acceptability. The control was preferred to pimentos containing either 90·9 or 136·5 mg. citric acid/4 oz. can ; those containing 90·0 mg. of acid and added sugar were preferred to those containing only added acid. (39 references.) E. M. J.

Varietal differences and storage changes in β-carotene content of six varieties of winter squashes. R. J. Hopp, S. B. Merrow and E. M. Elbert (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 568—576).—Varietal differences in the β-carotene content of squashes (pumpkins) were found, but in all varieties it increased during storage of mature fruits. L. G. G. WARNE.

Measurement of fibrousness of asparagus. A. Kramer, R. C. Wiley, B. A. Twigg, R. W. Decker and A. P. Sidwell (*Proc. Amer. Soc. hort. Sci.*, 1960, **76,** 382—388).—Bulk measurements of " fibrousness " (with a " shear press ") can be used to estimate variability within a sample. L. G. G. WARNE.

Effect of alternate freezing and thawing on the ascorbic acid content of frozen vegetables. G. J. Hucker and Ann Clarke (*Food Technol.*, 1961, **15,** 50—51).—The most significant factor in ascorbic acid loss is the time and temp. of holding during the thawed phase. Vegetables may be held from 4—8 h. after blanching and before freezing without appreciable loss in ascorbic acid. E. M. J.

Gelation of canned peas and pinto beans, as influenced by processing conditions, starch and pectic content. J. J. Powers, D. E. Pratt and J. B. Joiner (*Food Technol.*, 1961, **15,** 41—47).—The starch content, water-sol. pectic fraction and the Na hexametaphosphate-sol. fraction of the brine were each correlated with the degree of gelling or clumping of the canned product. Treatment of the peas and beans with pectolytic and amylolytic enzymes, $CaCl_2$ or Na hexametaphosphate influenced the extent of gelling or characteristics of the gel. (22 references.) E. M. J.

Statistical survey of pH variation and sodium content of tomatoes. H. W. Adams (*Food Technol.*, 1961, **15,** 16—18).—pH values of tomatoes (5000) were measured before and after peeling by steam or lye. The average pH value of lye-peeled is less than that of raw tomatoes. In steam-scalded tomatoes the Na values were much higher than in raw. Correlation between Na content and pH values was poor. Citric acid does not lower the pH of peeled tomatoes. E. M. J.

Non-alcoholic beverages

Technology of fruit concentration. D. Šulc and M. Ferić (*Fruchtsaft-Industr.*, 1960, **5,** 335—340).—Apparatus used in the prep. of concentrates of apples, grapes and stone fruits by vac. evaporation and methods of aroma recovery are described in detail. J. V. RUSSO.

Effects of fungicides on the flavour of fruits and syrups. A. Crang and G. M. Clarke (*J. Sci. Fd Agric.*, 1961, **12,** 227—234).—Captan (5 p.p.m.) was easily detectable in processed syrup especially in fruit-lacquered cans ; it was less easily detectable in fresh syrup but the syrups were liked less than controls ; metal cans showed corrosive action. Karathane in quantities up to 10 p.p.m. had little effect on the flavour of the syrup or puree and no apparent effect on the cans. Thiram was detected readily at 1 p.p.m. in canned syrup or puree ; it also attacked the cans. At 2 p.p.m. thiram increased rate of corrosion of the cans, caused distinct taint in canned but not in bottled syrups. In 5 years 1954—8 taint was observed, especially in canned fruit. E. M. J.

Moisture formation in dehydrated lemonade. W. K. Higby (*Food Technol.*, 1961, **15,** 47—49).—In absence of an in-package desiccant, moisture content increases during storage depending on the temp. as shown by browning and caking. The naturally present citric acid loses one carboxyl group giving a dicarboxylic acid and water, e.g., at 175°. This reaction is insignificant at temp. ranging from room temp. to 37°. E. M. J.

Tea, coffee, cocoa

Separation of glycerides of cocoa butter by paper chromatography. E. H. Steiner and A. R. Bonar (*J. Sci. Fd Agric.*, 1961, **12,** 247—250).—The separation of three mono-unsaturated glycerides of cocoa butter by reverse-phase chromatography and the location of their positions are described. R_F values for the three glycerides are given. The technique was applied successfully to illipé butter and cocoa butter " bloom " but not to butter fat and fats of the palm kernel type. (19 references.) E. M. J.

Milk, Dairy Products, Eggs

Reduction test in quality determination of milk. I. Mechanism of the methylene blue reduction. II. Specific reduction capabilities of various bacteria. M. Busse (*Z. LebensmittUntersuch.*, 1960, **113,** 453—464, 464—472).—I. In these tests the methylene blue (**I**) concn. was raised from 0·01 μmol./ml. to 0·5—3·0 μmol./ml. in buffered glucose media. Data are given for six species of lactic acid bacteria and stages in the glucose breakdown are discussed in detail. The **I** reduction was in fixed ratio to the acid formed but the ratio varied with species. (31 references.)

II. In optimal growing cultures, the specific **I** reduction capability

of *Bacterium coli* is considerably less than that of *Streptococcus lactis*. This is shown in the growth and log phase. With *B. coli* the reduction speed becomes a small % of max. value, while with *Str. lactis* the visible colour is reduced at nearly constant rate. In a mixture of the two organisms the streptococcus is determined by the duration of the **I** reduction. (17 references.) E. M. J.

[A] **Fatty acid composition of monoglycerides from lipolysed milk fat.** R. G. Jensen and G. W. Gander. [B] **Specificity of milk lipase towards primary ester groups of some synthetic triglycerides.** G. W. Gander and R. G. Jensen (*J. Dairy Sci.*, 1960, **43**, 1758—1761, 1762—1765).—[A] The monoglycerides of rancid milk (8 samples) are found (by gas–liquid chromatography of the corresponding allyl esters) to include all the acids associated with milk fat, but in widely differing proportions, viz., palmitic >31%, oleic and myristic >15%, and other acids <6%. Some of the allyl esters remain unidentified. (15 references.)

[B] In experiments with two synthetic fats, one with oleic acid in the middle and palmitic acid in the outer positions, and the other with the reverse structure, lipase is shown to hydrolyse the primary in preference to the secondary ester groups. (14 references.) P. S. Arup.

Further observations on the oxidation of milk fat and the lactoglobulin content of milk from Friesian cows of various milk yields. E. Pijanowski, S. Jasińska and A. Benedykcińska (*Roczn. Technol. Chem. Żywności*, 1960, **5**, 17—35).—Previous reports (cf. Pijanowski and Hyziak, *14th Int. Dairy Congress*, Rome, 1956, Vol. 1/1, p. 292, and Pijanowski, Habaj and Hyziak, *ibid.*, 1957, **1**, 35) that milk yields of the more productive Friesians generally contained more lactoglobulin, and that the fat was more liable to auto-oxidation on heating were confirmed. Cows on three farms were divided into lower and higher producing groups giving 2600 and 3900 l. respectively in a complete lactation season. The milk was sampled in 3—4 week intervals between late February and the end of June 1956. The milk fat oxidation was determined by the Lea peroxide and the 2-thiobarbituric acid tests, and a term "fat oxidation index" was introduced for mean peroxide no. after heating for 3, 4, 5, 6 and 7 h. The average figures obtained were: globulin content of milk 0·142, 0·151, fat oxidation index 5·29 and 7·8 for the two groups respectively. Additional tests of the cream enriched with globulins precipitated with $(NH_4)_2SO_4$ indicated a direct relation between the higher globulin content of milk and the increased oxidation of the fat. (10 references.) A. L. Grochowski.

Microscopical observation of effect of reagents on physical state of casein. N. King (*J. Dairy Res.*, 1960, **27**, 353—360).—The dispersing or coagulating effects on the casein miscella of separated milk or on those of acid or rennet coagula vary considerably with different reagents, with different concn. of the same reagent, and with the nature of the miscella. Urea has a dispersing effect on the coagula at 4M, and Na dodecyl sulphate at 0·1M; in the presence of EDTA (0·1M), however, urea (2M) has a distinct coagulating and fibre-forming effect on rennet coagulum (at pH 5·1); on acid coagula (at pH 4·7) the effect is merely granulating. Fibre formation (as that which occurs in cheddaring) probably depends on the destruction of Ca-bridges as well as on the cleavage of H-bonds and a limited range of pH. (10 references.) P. S. Arup.

Action of rennin and pepsin on β-casein: insoluble and soluble products. J. Cerbulis, J. H. Custer and C. A. Zittle (*J. Dairy Sci.*, 1960, **43**, 1725—1735).—After the treatment of β-casein (alone) with the enzymes (without coagulation), evidence of hydrolysis is observed by pptn. (partial) by adjustment to pH 4·7 or by the addition of 2% trichloroacetic acid. The treated β-casein is thus found to have been separated into sol. and insol. fractions, both of which prove to be of heterogeneous composition. The hydrolysing action of pepsin is more extensive than that of rennin. (10 references.) P. S. Arup.

Colloidal phosphate of milk. III. Nature of its association with casein. T. C. A. McGann and G. T. Pyne (*J. Dairy Res.*, 1960, **27**, 403—417).—The most noticeable effects of the removal of colloidal phosphate from milk (cf. *ibid.*, 9) are an approx. 30% increase in η, a greatly increased sensitivity to the coagulating effect of Ca salts, and (as shown by controlled renneting experiments) an apparent loosening of the association of the casein components; heat-stability is reduced, whilst stability to EtOH is unaltered. Reintroduction of colloidal phosphate causes a partial restoration only, of the original properties; heat-stability can, however, be wholly restored. These and other observations are considered to support the theory of the existence of chemical links between colloidal phosphate and casein in milk. (26 references.) P. S. Arup.

Estimation of the protein content of milk by dye binding with Buffalo Black. C. Vanderzant and W. R. Tennison (*Food Technol.*, 1961, **15**, 63—66).—Advantages over the dye Orange G are discussed. The accuracy of the procedure is comparable with that of the Kjeldahl method and protein values are not affected by refrigerated storage of the milk, with or without preservative, for 1 week; pasteurisation by holding or high-temp.–short-time method. The method should not be used for colostral milk or milk from cows with acute mastitis. (18 references.) E. M. J.

Binding of riboflavin and riboflavin phosphate by proteins of milk. A. Leviton and M. J. Pallansch (*J. Dairy Sci.*, 1960, **43**, 1713—1724).—The binding effects are limited to those of the component protein mol. of the milk proteins, and decrease so rapidly with temp. as to be ineffective at sterilising temp. There is no evidence of any special binding effect attributable to micellar structure. (17 references.) P. S. Arup.

Nitrogen fractions in centrifuge preparations from milk. C. H. Whitnah and R. Bassette (*J. Dairy Sci.*, 1960, **43**, 1731—1735).—The prep. from a single sample of milk, consisting of separated milk, wheys and sediments, are obtained with the use of a cream separator and two high-power centrifuges. A study is made of differences in results obtained by batch or continuous separation, and by variations in storage temp. before centrifuging and temp. during centrifuging. The prep., when analysed by the Aschaffenburg and Drewry method, showed variations in the distribution of the non-casein N fractions. P. S. Arup.

Effect of structure of some mercaptans on their amperometric titration with silver nitrate. K. Aibara, E. O. Herreid and H. K. Wilson (*J. Dairy Sci.*, 1960, **43**, 1736—1743).—Whilst titrations of nine of the compounds investigated give accurate results, the results for cysteine hydrochloride and penicillamine are ~25% too high. The discrepancies are explained by structural peculiarities giving rise to more than one ionisable form. Confirmation of the explanation is obtained by a study of the absorption spectra of the mercaptans and their Ag complexes. P. S. Arup.

Measurements of non-ionic iodine in milk of dairy cows following oral administration of labelled sodium iodide. A. Morgan (*J. Dairy Res.*, 1960, **27**, 399—402).—The iodide-I in milk can be completely removed by passage over a column of the exchange-resin Deacidite FF, pretreated with M-NaCl without affecting the content of non-ionic I. The amount of non-ionic I remaining in milk after 6 h. varies with individual cows from 1·2—14·6% of the total dose. P. S. Arup.

Absorptiometric determination of fluoride in milk. (*U.K.A.E.A.*, 1960, P.G. Rep. 141(S), 8 pp.).—The milk is curdled with rennet, made alkaline with NaOH, evaporated to dryness and the residue ashed at 600°. The fluoride is separated from this by steam distillation of aq. H_2SO_4 solution and determined absorptiometrically by its bleaching effect on aluminium Solochrome cyanine complex. A. C.

Identification of some volatile carbonyl compounds from non-fat [instant] dry milk. R. Bassette and M. Keeny (*J. Dairy Sci.*, 1960, **43**, 1744—1750).—The compounds developed in this product during storage have been isolated by a distillation process and separated by chromatography of their dinitrophenylhydrazones. The compounds presumably formed by the Maillard reaction include diacetyl, furfural, methyl-butanal and methyl-propanal. The compounds which (in addition to diacetyl) are the main contributors to the flavour are oxidation products including the C_6—C_{14} (except C_{11} and C_{13}) aldehydes; these are estimated to be present in amounts >2—3 μg. per l. (11 references.) P. S. Arup.

Analyses and quantitative determination of strontium-89 and -90 in milk powder. L. Jeanmaire and G. Michon (*Lait*, 1959, **39**, 369—381; *Rapp. Cent. Et. nucl. Saclay*, 1959, No. 1329).—Sr is separated from Ca by pptn. as NO_3^- and purified by ion-exchange. The radioactivity is measured before and after reaching equilibrium with ^{90}Y, which is precipitated as oxalate. Results obtained are in good agreement with those from conventional methods. A. C.

Fishy flavour in dairy products. III. Volatile compounds associated with fishy flavour in washed cream. [A] **Volatile compounds associated with painty and tallowy flavours in butterfat.** D. A. Forss, E. A. Dunstone and W. Stark (*J. Dairy Res.*, 1960, **27**, 373—380, 381—387).—III. The flavouring compounds developed at 2° in presence of Cu (5 p.p.m. as $CuCl_2$) and ascorbic acid (50 p.p.m.) are isolated and separated by gas-chromatography (cf. *ibid.*, 211, 373). The main contributors are the oily and the metallic fraction; the former contains a mixture of n-hexenal, n-heptenal and hex-2-enal, and the latter an unknown carbonyl compound; a fraction containing n-pentenal and pent-2-enal is painty in flavour, hept-2-enal mushroom-like, non-2-enal cucumber-like, whilst a fraction containing n-octanal, n-nonanal, oct-2-enal and hepta-2,4-dienal is tallowy. The chemical mechanism of flavour development is discussed. (19 references.)

[A] In comparison with the compounds associated with the fishy flavour (cf. previous abstract) those responsible for the tallowy

flavour caused by keeping butterfat containing dissolved Cu acetate (0·5 p.p.m. of Cu) at 45° during a few weeks contain relatively more of the C_{7-9} n-alkanals, heptan-2-one (present in traces in the fishy fractions), hept-2-enal and non-2-enal; the compounds causing the painty flavour (developed by the same treatment after 1—2 years) contain relatively more of n-pentenal and the C_{5-10} alk-2-enals; the total amounts of these compounds are ~10 and 100 times as great, respectively, as those associated with the fishy flavour; in both cases the content of the metallic-flavoured compound is insignificant. (12 references.) P. S. ARUP.

Detection of nitrous oxide in whipped cream. H. Sperlich (*Dtsch. LebensmittRdsch.*, 1960, **56,** 318—320).—The described method is based on thermal decomposition of the N_2O released from the cream on standing in an airtight container, and colorimetric detection of the resulting nitrous acid with sulphanilamide and *N*-(1-naphthyl)-ethylenediamine hydrochloride. No special apparatus is required for the test. E. C. APLING.

Effect of chemical additives on spreading quality of butter. I. Consistency of butter as determined by mechanical and consumer panel evaluation methods. J. G. Kapsalis, J. J. Betscher, T. Kristoffersen and I. A. Gould (*J. Dairy Sci.*, 1960, **43,** 1560—1569).—The construction and operation of an improved consistometer of the Heubner and Thomsen type (cf. *ibid.*, 1957, **40,** 834) are described. Results obtained for 109 samples show wide variations, a close relationship between spreadability (as mechanically determined by the knife) and hardness (as determined by the cutting wire), and satisfactory agreement with the findings of a testing panel. The important influence of the method of pasteurisation and of churning and storage temp. is demonstrated. (24 references.) P. S. ARUP.

Quantitative determination of *p*-hydroxybenzoic acid in cheese. F. Kiermeier and R. Jarczynski (*Z. LebensmittUntersuch.*, 1960, **113,** 370—374).—The naturally occurring *p*-hydroxybenzoic acid (**I**) in cheese was determined by boiling with HCl, filtering off the fat, extracting the filtrate with ether, separating the **I** paper-chromatographically and eluting, and measuring the colour produced with diazotised *p*-nitraniline. Amounts of **I** found in a soft cheese (Romadur) increased to about 0·4 mg./100 g. during ripening. The small amounts of naturally produced **I** in cheese would be negligible in the determination of **I** added as preservative. (11 references.) C. L. HINTON.

Factors affecting the activity of spray-dried cheese culture. C. W. Sapp and T. I. Hedrich (*Mich. agric. Exp. Sta. quart. Bull.*, 1960, **43,** 96—104).—The cultures dried by ordinary spray-drying equipment retained sufficient activity to clot and coagulate high-temp. pasteurised skim milk, after a 1% inoculation, in 16 h. at 75°F. Optimum drying was obtained with the outlet temp. of the drier at 135—165°F. A. G. POLLARD.

Heat resistance of three strains of psychrophilic organisms added to skim milk for cottage cheese manufacture. R. A. Chaudhary, S. L. Tuckey and L. D. Witter (*J. Dairy Sci.*, 1960, **43,** 1774—1782).—These strains of *Pseudomonas viscosa*, *P. fluorescens* and *P. fragi* do not survive normal pasteurisation temp. or the conditions prevailing in the curd during manufacture. (20 references.) P. S. ARUP.

Volatile compounds in New Zealand Cheddar cheese and their possible significance in flavour formation. III. Time of first appearance of volatile carbonyl compounds during ripening. R. J. Harvey and J. R. L. Walker (*J. Dairy Res.*, 1960, **27,** 335—340; cf. *ibid.*, 1959, **26,** 265).—MeCHO, $COMe_2$ and butan- and pentan-2-one can be detected in steam distillates of 1-day-old cheese; heptan-2-one appears after 2—4 weeks, nonan-2-one after 20—24 weeks, and undecan-2-one after 36 weeks. The content of these compounds increases steadily with age. The development of Cheddar flavour is especially associated with the appearance of the higher ketones. P. S. ARUP.

Properties and food uses of ducks' eggs. M. B. Rhodes, J. L. Adams, N. Bennett and R. E. Feeney (*Poultry Sci.*, 1960, **39,** 1473—1478).—The eggs of the Khaki Campbell duck compared favourably with chicken eggs with respect to the flavour of cooked eggs and cakes made from the eggs. Duck eggs were superior to chicken eggs with respect to greater stability to storage deterioration, less evident chalaza, reduced darkening around the yolks of hard-boiled eggs, and less volatile S. Disadvantages of duck eggs over chicken eggs are the firm white and the tough shell membrane. A. H. CORNFIELD.

Milk powder. Aplin & Barrett Ltd. (Inventor: H. B. Hawley) (B.P. 801,740, 3.12.54. Amended, 17.2.60).—A two-stage process for the spray-drying of whole or skim milk comprises preheating to 77—99° then further preheating during >3 sec. at 130—150°, prior to spray-drying (cf. J.S.F.A. Abstr., 1960, i, 92). F. R. BASFORD.

Edible Oils and Fats

Chemical investigation of seeds of selected tropical plants. I. Component acids of the fats or oils. A. Mackie and D. G. Mieras (*J. Sci. Fd Agric.*, 1961, **12,** 202—205).—The following seeds were studied: the Malayan Pinang mabuk, a variety of betel nut *Areca catechu*, Palmae and the West African seeds, *Monodora myristica*, *Xylopia aethiopica*, *Celosia argentea*, *Anogeissus schimperi*, *Mangifera indica* and *Carapa procera*. Isolation of any vermicidal principle was considered, since the seeds were used by natives as anthelmintics. Data are given on fatty acid determinations by gas chromatography. The contents of component acids are compared; e.g., members of the same family Anonaceae, *X. aethiopica* contains ~10 times the amount of palmitic acid and $\frac{1}{16}$ the amount of linoleic acid as does *M. myristica*, but about twice the amount of palmitic acid and >0·25 the amount of linoleic acid present in other members of the Anonaceae. E. M. J.

Membranes protecting oil emulsions in protein solutions. J. M. Martínez Moreno, C. Gómez Herrera, E. Márquez Delgado, C. Janer del Valle and L. Duran Hidalgo (*J. Amer. Oil Chem. Soc.*, 1960, **37,** 582—587).—Electron-microscope and -diffraction studies of the membrane structure and its ability to adsorb selectively heavy metals are described, with particular reference to olive oil in olive juice. There is evidence of the existence of a single spot or "pole" in the globular membrane, which is probably associated with emulsion breakdown. Emulsions prepared with ultrasonic waves were shown to form subdivided droplets or "clusters." P. M. KINGSTON.

Fatty acid composition of the depôt fats of the kiwi (*Apteryx australis mantelli*). F. B. Shorland and J. P. Gass (*J. Sci. Fd Agric.*, 1961, **12,** 174—177).—The fatty acid composition of the depôt fats of kiwi is similar to that of other land birds except for the occurrence of substantial amounts of C_{20} highly unsaturated acids. (20 references.) E. M. J.

Selective hydrogenation of vegetable and animal unsaturated oils and fats. Buss A.-G. (B.P. 834,817, 11.6.56. Switz., 14.6.55).—In the process where a mixture of the material to be hydrogenated in the liquid state and a hydrogenation catalyst is circulated in a closed cycle through a vessel, H_2 is passed into the circulating line at a point on the pressure side in amount >0·5% by wt. (of the wt. of the material to be hydrogenated), and the mixture of material to be hydrogenated and the catalyst mixed with the H_2 on the pressure side of the circulating means. The apparatus is claimed. E. ENOS JONES.

Meat and Poultry

Enzymic changes in lambs' liver during storage. I. D. N. Rhodes and C. H. Lea. **II. Activation of autolytic enzymes by freezing.** D. N. Rhodes (*J. Sci. Fd Agric.*, 1961, **12,** 211—224, 224—227).—I. Rates of autolytic degradation of phospholipids, neutral lipids and protein were measured during storage at 37°, 15°, 1°, −10° and −20°, and at 15° and 1° after frozen storage at −10° or −20° for 8 or 14 weeks. In fresh liver, rate was 13 times faster at 37° than at 15° and ~3·5 times faster at 15° than at 1°. After storage for 24 weeks at −10° and −20° change was hardly detectable. When this stored liver was then thawed at 1° the activating effect of freezing on the hydrolytic enzymes was much less marked than when post-thawing storage was at 15°. In degradation of glycerophospholipids removal of two fatty acid residues, followed by breakdown of water-sol. phosphate ester moiety to orthophosphate, occurs. Neutral fats are rapidly hydrolysed, indicating lipase activity. Fresh liver was acceptable for up to 10 days at 1°, or 1—2 days at 15°, and no significant deterioration was detected organoleptically after storage for 25 weeks at −10° or −20°. (20 references.)

II. The apparent acceleration (3—4-fold) of autolysis in liver after freezing and thawing is mainly the result of elimination of the lag phase which, in fresh material, precedes the onset of the rapid phase of enzymic degradation of lipids and protein. Freezing at −10° is complete within 3 days and at −2° within 5 days. Activity is not enhanced by further frozen storage at −10° or −20° for periods up to 14 weeks. The formation and growth of ice crystals during freezing could cause breakdown of the gross physical organisation of the tissue, dehydration of cellular constituents, denaturation of proteins and disruption of lipoprotein complexes. Termination of the lag phase might be due to release of enzymes by digestion of the membrane by the indigenous protease. (10 references.) E. M. J.

Water binding capacity of fresh pork. I. Influence of sodium chloride, pyrophosphate and polyphosphate on water absorption. II. Influence of phosphates on fat distribution in meat products.

III. Influence of cooking temperature on the water binding capacity of lean pork. P. Sherman (*Food Technol.*, 1961, **15**, 79—87, 87—89, 90—94).—I. Fluid retention at 0° (**I**) from aq. NaCl and other neutral salt solutions depends on the degree of ion absorption and correlates well with anion absorption. With phosphate solutions **I** shows a significant statistical correlation with pH of the aged solution–meat mixtures. pH also affects the degree of solubilisation of (acto)myosin from meat proteins. At 100° anions and cations are released by meat but anions are still preferentially retained from neutral salts. In presence of phosphates, fluid retention at 100° is related to concn. of (acto)myosin going into solution at 0° during ageing of the solution–meat mixtures. The gel of denatured protein extends throughout the meat mass and retains moisture. These effects are influenced by solution–meat ratio. (33 references.)

II. In sausages soap formation is promoted when added phosphate is very alkaline, and the fat contains a high proportion of free fatty acid. If NaCl is present any soap that is formed will be "salted out." Since free fatty acid content is extremely low, soap formation, even in absence of NaCl, is at too low a level to emulsify the fat. (19 references.)

III. On heating ground pork mixed with aq. NaCl or solutions of alkaline phosphate at 25—100°, the amount of fluid retained by the meat shows an overall decrease with increase in temp. Variations in fluid retention on heating are explained in terms of colloidal transformations that occur in the sol. meat proteins. The most important factor is solubilisation or swelling of the proteins, particularly (acto)myosin, within the meat prior to heating. This process is influenced by pH, time and temp. of the initial ageing period, additive concn., and, for alkaline phosphates, an additional factor, ability to split the bond between actin and myosin in (acto)myosin. E. M. J.

Relation of marbling, cooking yield and eating quality of pork chops to backfat thickness on hog carcasses. M. O. Murphy and A. F. Carlin (*Food Technol.*, 1961, **15**, 57—63).—The average flavour scores for chops summarised from recorded data indicated the similarity in flavour regardless of backfat thickness or of marbling. (10 references.) E. M. J.

Nitrate and nitrite metabolism in a bacon-curing brine and their relation to the bacterial population. B. P. Eddy and A. G. Kitchell (*J. Sci. Fd Agric.*, 1961, **12**, 146—152).—The effects of salt concn., temp. and bacterial population density on rates of metabolism of nitrite and nitrate were studied in a bacon-curing brine in which NO_2^- and NO_3^- were initially absent. The most rapid changes take place at 10° in 20% salt and the least rapid at 5° in 25% salt. There is an apparent lack of correlation between increases in bacterial no. and rate of metabolism. Increases in metabolic activity are more closely related to the total than to viable counts. E. M. J.

Fat content of meat sausages. K. Philippi and L. Bertling (*Dtsch. LebensmittRdsch.*, 1960, **56**, 320—323).—The fat contents of some 600 samples of various types of sausages from different areas of Germany, analysed during 1956—8, are reported and their significance is discussed. The mean results for different sample groups varied only from 27·9% to 32·3%, but the range of fat contents of individual samples was from <20% to >40%. E. C. Apling.

Fish

Storage changes in frozen fish. A comparison of objective and subjective tests. K. Andersson and C. E. Danielson (*Food Technol.*, 1961, **15**, 55—57).—Herring fillets were dipped in 5% ascorbic acid, frozen and analysed periodically by the 2-thiobarbituric acid (TBA) method and organoleptic tests. Changes in taste corresponded with chemical changes shown by the TBA tests; the fillets were palatable for 11 months. Untreated samples were rancid after storage for 2 months. E. M. J.

Spoilage of Pacific coast rockfish. I. Spoilage in ice storage. J. Liston, J. G. Chapel and J. A. Stern (*Food Technol.*, 1961, **15**, 19—22).—The pattern of spoilage of *Sebastodes alutus*, *S. melanops* and *S. pinneger* as indicated by the mathematical relationship between organoleptic scores and chemical indexes is similar to that reported for cod and haddock. E. M. J.

Spices, Flavours, etc.

Citrus essential oils. I. Evaluation of natural and terpeneless lemon oils. C. A. Slater (*J. Sci. Fd Agric.*, 1961, **12**, 257—264).—Samples were examined by a combination of absorption and gas chromatography and i.r. spectroscopy. Natural lemon oil contains ≮7 terpenes, 9 sesquiterpenes and ≮24 oxygenated compounds. By examination of the oxygenated and hydrocarbon fractions the genuine quality and freshness of the oil were determined but organoleptic testing is of additional value. (13 references.) E. M. J.

Combined paper-chromatographic and polarographic determination of vanillin and ethyl-vanillin in foods. II. H. Woggon, K. Rauscher and U. Köhler (*Nahrung*, 1960, **4**, 374—389).—Vanillin (**I**) and ethyl-vanillin (**II**) can readily be separated on paper by known methods, without interference by sugars or water-sol. colouring matters. Measurements of the test-samples of extracts of **I** and **II** from foods (made with EtOH or Et_2O) should be made with a micrometer-pipette. The spots (visible in u.v. light) are eluated by a 0·5M solution of semicarbazide hydrochloride in 0·1N-HCl, which serves as the base solution for the (separate) polarographic evaluation of **I** and **II**; for **I**, $E_{\frac{1}{2}} = 0{\cdot}76$ V, and for **II**, 0·75 V. The extraction of **I** and **II** from starchy products is accomplished by a short treatment with conc. aq. NaOH which is then made acid and extracted with Et_2O. For **I** (5—15 μg./ml.) the average error is ±2%, and for **II** (2—5 μg./ml.) ±4%. For concn. <2 μg./ml., the error is ±7%. (17 references.) P. S. Arup.

Preservatives

Possibilities of optimum utilisation and increased efficiency of limited amounts of chemical preservative additives. M. L.-v. Schelhorn (*Nahrung*, 1960, **4**, 332—346).—A review covering the influence of initial infections on preservative requirements, the use of mixed preservatives, the combination of physical and chemical methods of preservation and the use of chemically impregnated packing material. (22 references.) P. S. Arup.

Food Processing, Refrigeration

Food preservation by ionising radiation. H. W. Nelson (*Battelle Tech. Rev.*, 1961, **10**, No. 1, 8—15).—A diagrammatic food-irradiation line is presented, a linear accelerator being employed to irradiate food cartons from above and below as they move along the conveyor. The effect on meats, poultry, seafoods, vegetables and fruits is summarised together with a brief review on odour, inhibition of sprouting, rain disinfestation, etc. Comparative costs are given for irradiation (electron acceleration and ^{137}Cs at 10^6 and 3 Mrad.), thermal and freezing methods. C. V.

Dehydrofrozen apples: recent developments in processing methods. M. E. Lazar, E. O. Chapin and G. S. Smith (*Food Technol.*, 1961, **15**, 32—36).—Through-flow drying was substituted for cross flow. Three piece sizes and three different varieties were processed under various conditions. Advantages of using an optional blanching step after drying are discussed and a procedure is given for a blanched non-sulphured product. (12 references.) E. M. J.

γ-Radiation in the control of decay in strawberries, grapes and apples. L. Beraha, G. B. Ramsey, M. A. Smith, W. R. Wright and F. Heiligman (*Food Technol.*, 1961, **15**, 94—98).—Irradiation at from 100,000—300,000 rep substantially reduced grey mould *Botrytis cinerea* and *Rhizopus* rot of strawberries during storage at 75°F for 3 days or 41° for 10 days. Doses of 300,000 rep prevented decay of berries stored for 8 days at alternating temp. of 41°F and 75°F; injury to fruit was observed after storage for 4 days and severe injury after 8 days. Under these conditions doses of 200,000 rep prevented decay for 7 days and 100,000 rep for 6 days without visible injury to berries. At 75°F a dose of 50,000 rep did not reduce growth of *Penicillium expansum* in Jonathan apples regardless of age or infection. Tokay grapes inoculated with *B. cinerea* remained free of decay for 4 days at 75°F after 200,000 rep. E. M. J.

Analysis of headspace gases in canned foods by gas chromatography. D. C. Vosti, H. H. Hernandez and J. B. Strand (*Food Technol.*, 1961, **15**, 29—31).—A method is described of determining microquantities of N_2, O_2, CO_2 and H_2 by incorporating a two-stage column and two thermal conductivities detectors in a gas–solid chromatographic separation apparatus. Argon is used as the carrier gas and silica gel and mol. sieve as the adsorbents. (10 references.) E. M. J.

Gas chromatography of carbon dioxide, hydrogen, oxygen and nitrogen in processed foods. B. S. Luh and M. S. Chaudhry (*Food Technol.*, 1961, **15**, 52—54).—Headspace gases of processed foods were examined by a silica gel–mol. sieve column with A or He as a carrier gas. In canned whole apricots swelling occurred and H_2 accumulated in the head space; in canned tomato serum, a Maillard-type of reaction occurred and CO_2 accumulated. O_2 in the head space is deleterious to processed foods containing unsaturated oils and fats, e.g., salad oil, French dressing and instant mashed pototoes. (13 references.) E. M. J.

Liquid nitrogen freezer adapted to use in the production line. Anon. (*Quick Froz. Fds*, 1960, **22**, No. 11, 31, 157—158).—Up to the present liquid-N_2 has been used almost exclusively for transportation problems. A machine (16 × 8 × 8 ft.) has been designed to fit into the last phase of the machine production line; it has a capacity

of 120 packages/min. The packages arrive at 160—180°F, are placed in wire baskets and sent to a cradle-type conveyor which passes through a gentle stream of liquid N_2 (**I**). The first exposure freezes the top layer and prevents any subsequent spoilage. Moving from the shower area the packages are immersed in a pool of **I** at −320°F and the overall speed can be regulated to type, size and density of the product. The unit works as efficiently with raw materials as with large cartons. A re-liquifier is included so that **I** can be re-cooled with air removal for re-use. This method is considered competitive with normal refrigeration methods as regards transportation. C. V.

Preservation of meat products. Zwanenberg's Fabrieken N.V. (B.P. 835,984, 5.9.58. Neth., 27.9.57).—A process for the preservation of non-homogeneous meat products (fish, pork, beef, poultry) comprises placing the product between at least two pairs of electrodes (such that there is continuous contact between the meat and the electrodes), then charging the latter with low-frequency alternating voltage to heat the meat to preservation temp. under such conditions that the power supply of each pair of electrodes is a function of the difference in temp. between that pair of electrodes and the average temperature of all the electrodes present, and that the power supply is highest on the pair of electrodes between which the lowest temp. is measured. Apparatus is claimed. F. R. BASFORD.

Packaging

Frozen foods in boilable bags. F. R. Fleischman (*Quick Froz. Fds*, 1960, **22**, No. 8, 221—222, 224, 226, 229—230, 232, 238).—The history from 1940 is sketched and the future reviewed. The advantages and disadvantages of transparent (plastic) or opaque (Al) packings are discussed. C. V.

Packaging and storage of deep-fat-fried vegetables. B. S. Bhatia, J. V. Prabhakar and Girdhari Lal (*Indian J. appl. Chem.*, 1960, **23**, 73—80).—The safe limit of moisture content of deep-fat-fried beans, peas and raw banana chips is 5% at R.H. 60%. Storage at room temp. is possible for at least a year and may be prolonged by using an antioxidant with the salt used for dusting. If this is not used, refined coconut oil gives better results than hydrogenated groundnut oil. The colour is stable, except with banana; in this case it is improved by sulphite steeping prior to frying. A. C.

Testing of paper and other sack materials for penetration by insects which infest stored products. P. M. Davey and T. G. Amos (*J. Sci. Fd Agric.*, 1961, **12**, 177—187).—Seven types of sack materials used for storage of maize, groundnuts, cocoa beans, etc., and nine species of insects were studied. Detailed findings are presented and recommendations include: use of multi-ply Kraft paper as sacks or liner to jute sacks against *Tribolium castaneum* and *Sitophilus oryzae*; use of 0·005-in. polyethylene as a sack liner for malting barley; fumigation of infested produce immediately on filling the sacks; patch-sticking after insertion for sampling or handling; and for certain insects the use of a combined mechanical barrier and an insecticide deposit. (14 references.) E. M. J.

Polyethylene liners and fungicides for peaches and nectarines. D. A. Luvisi and N. F. Sommer (*Proc. Amer. Soc. hort. Sci.*, 1960, **76**, 146—155).—Even low-density polyethylene film when used as box lining for peaches and nectarines is not sufficiently permeable to prevent CO_2 accumulation and the development of alcoholic flavours at moderate temp. Dipping in aq. dehydracetic acid and Na *o*-phenylphenate controlled post-harvest infection by *Monilinia fructigena* and *Rhizopus stolonifer*. L. G. G. WARNE.

Oxygen-scavenging packet for in-package deoxygenation. D. Scott and F. Hammer (*Food Technol.*, 1961, **15**, 99—104).—Developed for use with dehydrated foods, shape of packet, container, location and particle size (for solid particles) are shown not to affect rate or completeness of O_2 removal. Spray-dried products require more time to deoxygenate than foam or drum-dried products. The stoicheiometric relationships and implications of OxyBan as a substitute for vac. and gas packing are discussed. (19 references.) E. M. J.

Miscellaneous

Nutrition, proteins, amino-acids, vitamins

Investigation of mess-rations with regard to nutritional physiology. E. Wunderlich (*Nahrung*, 1960, **4**, 324—331).—A report on conditions obtaining in four large-scale kitchens in Germany with respect to the kcal.-balance between fat, protein and carbohydrates. Excesses of fat, sufficiencies of protein and deficits of carbohydrates are recorded. P. S. ARUP.

Effect of dietary fat on serum lipin fractions. J. R. Gilbertson (*Dissert. Abstr.*, 1960, **21**, 746).—The substitution of safflower oil for butter in adult human diet causes significant decreases in cholesterol (free and esterfied), phospholipins and (especially) in the triglycerides. No changes are observed in the free fatty acid content. Cholesterol linolenate disappears after changing over to the vegetable oil diet. P. S. ARUP.

Nature of fatty acids of human depot fat. J. T. Dickman (*Dissert. Abstr.*, 1960, **21**, 745—746).—Analytical data for the fractional distillates of the Me esters of the fatty acids (previously fractionally crystallised) suggest the presence of isomers of octadecanoic acid having at least one double bond of *trans*-configuration, and of dienoic acids which are mostly non-conjugatable and of *cis-cis*-configuration, representing 1·3% of the total. Gas-chromatographic analysis of fractions of the esters (obtained by crystallisation followed by distillation) point to the occurrence of n-tridecanoic acid (0·003—0·02%), n-pentadecanoic acid (0·20—0·56%), n-pentadecenoic acid (0·03—0·08%), n-heptadecanoic acid (0·21—0·37%) and n-heptadecenoic acid (0·1—0·5%). Samples representing diseased conditions show wide variations in polyene-acid content. P. S. ARUP.

Heated fats. II. Nutritional properties of heated cottonseed oil and of heated cottonseed fractions obtained by distillation and urea adduct formation. L. Friedman, W. Horowitz, G. M. Shue and D. Firestone (*J. Nutr.*, 1961, **73**, 85—93).—Cottonseed oil heated 190 h. in air at 225° was fractionated into linear and non-linear monomers, dimers and high polymers by molecular distillation and urea adduction. This heated oil when fed to rats in a 6-month study or in a 10—12 day "kcal. assay" was found to have decreased in nutritional value as regards growth and gave rise to enlarged livers. The cause of these changes was a fatty acid which had lost its capacity to complex with urea. It was suggested that the quant. determination of this may be a useful test for judging the acceptability of used fats in commercial cooking or specific industrial operations. (21 references.) C. V.

Protein and lipid constitution of Pakistani pulses. B. E. Baker, J. A. Papaconstantinou, C. K. Cross and N. A. Khan (*J. Sci. Fd Agric.*, 1961, **12**, 205—207).—Nineteen fatty acids were determined by gas chromatography in the light petroleum extracts of five common pulses. Data are given on the effects of extraction with water and salt concn. (0·005—1·0N) on peptisation of protein of these pulses. With mash, mung, masur and gram two minima were observed from the plotted curves, but with lobia only one minimum was observed. (15 references.) E. M. J.

Nutritive value of fresh fish, conserved fish and fish meal. W. G. Jaffé (*Z. LebensmittUntersuch.*, 1960, **113**, 472—479).—Samples of fish and fish products (28) and of fish meal (10) were analysed for water content, ether extract, raw protein, ash, vitamin and amino-acid contents and thiaminase activity. The effect of fish meal on the biological value of wheat flour and white bread was studied. The significance of fish meal for enriching cereal proteins and its compatability is shown by theoretical calculation and rat tests. Fish meal was added to bread without affecting baking properties or taste. (16 references.) E. M. J.

Large-scale production of protein from leaf extracts. J. E. Morrison and N. W. Pirie (*J. Sci. Fd Agric.*, 1961, **12**, 1—5).—Fresh leaves (e.g., cereals, mustard, clover, lucerne) are pulped with water or alkali and the juice is pressed out. Starch and small amounts of other non-protein material are removed and the protein is coagulated with steam, filtered and washed with water at pH 3·5—4. The final product is put into moulds 3 in. × 10 in. × 10 in., placed in a cheese press overnight to raise the dry matter content to 30—40%, wrapped in polyethylene film and stored at −10°. Flavour and physical properties do not alter on storage for a few weeks but the texture becomes gritty after months of frozen storage. The effects of drying, especially freeze drying, leaf protein are discussed. N content is 10—12% (e.g., for cereals); 10% (grass); 8% (bracken); (19 references.) E. M. J.

Leaf protein concentrates. I. Effect of source of raw material and method of drying on protein value for chicks and rats. J. Duckworth and A. A. Woodham. **II. Value of a commercially dried product for newly-weaned pigs.** J. Duckworth, W. R. Hepburn and A. A. Woodham (*J. Sci. Fd Agric.*, 1961, **12**, 5—15, 16—20).—I. The nutritive value of protein of leaf-protein concentrates made from mixed grasses, barley, kale, rye, tares was uniformly high, the gross protein values being 71, 77, 75, 82, 82 respectively compared with that of soya-bean meal, 74. Gross protein value was not affected by drying at low temp. but drying at 100° in a slow current of air reduced the value of (barley) concentrate to 39. (22 references.)

II. The value of a (wheat) leaf-protein concentrate (**I**) dried by a commercial process (not causing decrease in nutritive value, as

measured in chicks and rats) was tested in newly-weaned pigs. When ~7% of **I** was included in the diet (barley meal–millers' offals, 2 : 1) the rate of growth and efficiency of feed utilisation of the pigs were similar to those fed a standard diet containing 7 or 8% of white fish meal ; when 10% was included, efficiency of food utilisation was improved ; when 4—5% of **I** was included and the remainder of the supplementary protein made up from groundnut meal growth rate and efficiency were improved. E. M. J.

Extraction of protein from leaves of plants growing in Ghana. M. Byers (*J. Sci. Fd Agric.*, 1961, **12,** 20—30).—The amount of extractable protein from 60 species of leaves was determined. The fresh leaves were minced and the pulp was squeezed through cotton cloth. The results are classified with regard to the extractability and quality of the protein, the content of which varied from 15 to 70%. Legumes are a valuable source of easily extractable and good-quality protein as shown by the eight species examined. More protein was extractable from some of the more common weeds than from leaves of crops already grown for food. Extraction of some leaves was difficult as they contained mucilage or much fibre, but different extraction techniques might overcome the mechanical difficulties involved. (11 references.) E. M. J.

Problems in the prediction of protein values of diets. D. S. Miller and P. R. Payne (*Brit. J. Nutr.*, 1961, **15,** 11—19).—A method is presented which enables the protein-value (**I**) of diets to be predicted from analytical data. Dietary values are shown in terms of the proportion of energy in the diet contributed by the protein. The net dietary-**I** required for maintenance is 4% of the total kcal. and the % of protein kcal. is given by P_m = (400/score) where the score is a satisfactory estimate of the net protein utilisation (standardised). For diets having a n.d.—p.v. (net dietary—**I**) >4% of the kcal. (e.g., when the score × P > 4), the equation at different protein levels is n.d.—**I** = P × score $[(54 - P)/(54 - P_m)]$ where P = % protein kcal. in the diet and P_m = % energy in the diet supplied by the protein. (14 references.) C. V.

Evaluation of protein quality in mixed foods. J. M. McLaughlan and A. B. Morrison (*Canad. J. Biochem. Physiol.*, 1960, **38,** 1378—1380).—In the mixtures half the protein was supplied (to rats) by bread and the other half by bread, casein, whole egg powder, fish flour, cooked beans protein, cereal + milk (1 : 4), Cheddar cheese or oatmeal. For mixtures of foods in which cereal products contribute about half or more of the protein, the lysine content is a reliable guide to the nutritional value of the protein mixture. E. M. J.

Experimental approaches to preferential hydration of proteins. D. J. Cox (*Dissert. Abstr.*, 1960, **21,** 745).—The relative advantages and limitations of three methods for determining the hydrodynamic *d* of pptd. proteins + water and salt (as a measure of preferential hydration) are examined. A modification of the "banding technique" of Meselson *et al.* allows of the detection of *d*-heterogeneity in ppt. which are mixtures. Preferential hydration values for a no. of proteins are 0·17—0·35 g. of excess water per g. of protein ; the value for bovine serum albumin in $(NH_4)_2SO_4$ or LiCl is 0·5—0·8 g. per g. The value for ribonuclease (0·35 g.) is the same between 10 and 40°, and pH 4·7—9·2. The reasons for the observed variations in the apparent *d* of pptd. bovine serum albumin with temp., pH, and concn. and kind of salt are considered not to be necessarily due to variability of preferential hydration.
P. S. Arup.

Splitting of amide and peptide bonds of dinitrophenyl peptides with anhydrous trifluoroacetic acid. E. Taschner and A. Chimiak (*Bull. Acad. polon. Sci., chim.*, 1960, **8,** 423—427).—When heated with anhyd. trifluoroacetic acid at 100° 2,4-dinitrophenyl peptides are cleaved to give dinitrophenylamino-acids and *N*-trifluoroacetyl amino-acids, which may be identified chromatographically. Traces of free amino-acids are also present. The reagent may be used for degradation of *N*-protected peptides and characterisation of *N*-terminal amino-acids. (14 references.) (In English.) A. C.

Milk substitutes of vegetable origin. II. The effect of fortification with DL-methionine on the nutritive value of spray dried powder obtained from a blend of soya-bean and groundnut milks. S. R. Shurpalekar, M. R. Chandrasekhara, N. L. Lahiry, M. Swaminathan, K. Indiramma and V. Subrahmanyan (*Ann. Biochem.*, 1960, **20,** 145—156).—Soya-bean, groundnut and modified cow's milk powders and their blends with or without methionine supplement were fed to albino rats ; the growth rate, composition of blood, liver and carcass and the protein efficiency ratio (PER) of the rats was then determined. Addition of methionine to vegetable milk powders increased the growth rate almost to that produced by modified cow's milk and also increased the PER. No significant differences in the content of haemoglobin, red blood cells, moisture and fat of liver or carcass was observed between rats fed different powders. (17 references.) S. A. Brooks.

Separation of amino-acid mixtures on sulphonated polystyrene resin-loaded papers. C. S. Knight (*Nature, Lond.*, 1960, **188,** 739—740).—The separation of 16 amino-acids on papers loaded with different resins is described, the best resin being "Zeocarb 225 (W.R. 1·5—2·0)." Suppression of the ion-exchange characteristics enables a two-dimensional technique to be used which combines ion-exchange and partition techniques. Excellent resolution is obtained in a single chromatogram A. C.

Unclassified

Pure and applied analytical chemistry and bromatological analysis. J. A. Gautier (editor) (Masson & Cie, 1960, Series 8, 148 pp.).—Reviews. **Gas-phase chromatography : principles and applications.** P. Chorin, pp. 6—29. **Possibilities of microbiological assay.** A. Desvignes, pp. 32—56 (89 references). **Bacteriological analysis of sugars and syrups.** P. Devillers, pp. 58—75. **Analytical problems arising from the use of pesticides in agriculture.** G. le Moan, pp. 78—118 (232 references). **Determination of *C*-methyl groups and identification of alkoyl groups in organic molecules.** F. Percheron, pp. 120—140 (41 references). H. S. R.

Microbial spoilage of canned food. III. Isolation of thermophilic cocci from an imported canned fish. G. Rangaswami and R. Venkatesan (*Proc. Indian Acad. Sci.*, 1960, **51B,** 264—269).—*Streptococcus thermophilus* Orla-Jensen was isolated from a can of "herrings" in tomato juice imported from Scotland. The spoilage effect of the organism on the can contents is described. The optimum pH for growth and the thermal death-point were determined.
S. G. Ayerst.

Amphoteric polyelectrolytes. Monsanto Chemical Co. (B.P. 833,459, 7.9.56. U.S., 8.9. and 16.11.55).—The products are made by interaction of an alkali-sol. polymer containing CO_2H groups (e.g., a copolymer of maleic anhydride or acid with a vinyl compound) with a substituted hydrazine (I—3 alkyl, aryl, alkaryl or aralkyl groups) or a dialkylformamide complex of a substituent hydroxylamine. The compounds are useful as flocculating agents for beer, soft drinks, in sugar-refining and sewage treatment and also as soil conditioners. H. S. R.

3.—SANITATION

Entry of fungi and chemical substances in solution into mature cereal grains. F. Caldwell (*J. Sci. Fd Agric.*, 1961, **12,** 169—174).—The literature is reviewed and discussed with reference to the author's own work on the influence of the semi-permeability of mature cereal grains on the entry of fungi and chemical compounds in solution. The viability and response of the grain to treatment with fungicides, e.g., Cu fungicides, in which the fungi are injured and the plant is stimulated are considered. (28 references.) E. M. J.

Removal of *Staphylococcus aureus* from milk-contact surfaces by ultrasonic cleaning. E. B. Masurovsky and W. K. Jordan (*J. Dairy Sci.*, 1960, **43,** 1545—1559).—Exploratory investigations are described of the influence of various factors on the efficient removal of ^{32}P-labelled *S. aureus* from (experimentally) milk-soiled specimens of dairy plant. Radiological methods for evaluating the efficiency of cleaning are compared. The efficiencies of different types of cleaning solution are studied. Stress is laid on the importance of the relative configurations of the cleaning bath and the specimens treated, the nature of the deposits, and the necessity for their complete removal by every treatment. (18 references.) P. S. Arup.

Iodine disinfectants. J. L. Wilson, W. G. Mizuno and C. S. Bloomberg (*Soap, N.Y.*, 1960, **36,** No. 12, 100, 105—107, 137, 141, 143 ; 1961, **37,** No. 1, 105, 109, 111—112, 116 ; No. 2, 121—122, 124).—A brief review of the history and use of I and I-compounds is given and the action is compared with other compounds and proprietary preparations. The use in food processing plants, dairy farms, etc., is briefly discussed and equipment is described for the automatic injection of correct doses into flowing water for spraying, flushing, etc. Many references are included. C. V.

Genetics of house fly (*Musca domestica*) resistance to malathion. R. L. Harris, S. Wearden and C. C. Roan (*J. econ. Ent.*, 1961, **54,** 40—45).—A preliminary study. (14 references.)
C. M. Hardwick.

Toxicity to house fly larvae of droppings from chicks given Dipterex-treated water. M. Sherman and E. Ross (*J. econ. Ent.*, 1960, **53,** 1066—1070).—Solutions of Dipterex (30 p.p.m.) in acidified or distilled water administered to chicks resulted in persistently high larval mortality in the droppings. Tap water solutions of Dipterex produced droppings which lost their activity in a few days. At these dosages gain in wt. of and water consumption of chicks were

unaffected. Dipterex was excreted as a more toxic product but administered DDUP was excreted as a less active larvicide.
C. M. HARDWICK.

Biology and control of little house fly, *Fannia canicularis*, in Massachusetts. P. C. Steve (*J. econ. Ent.*, 1960, **53**, 999—1004).—Rearing techniques, life history, field recognition and oviposition preferences amongst different manures are discussed. Adult flies preferred a cool environment and were positively phototropic and the larvae negatively phototropic. Populations were much higher on poultry farms employing a pit system than on those using an open floor arrangement. Seasonal distribution is discussed. Fly cords containing parathion + diazinon gave reductions to a low level within 4 days on two farms and 12 days on another. No population build-up occurred in the next 120 days. *Musca domestica* needed additional treatment with malathion bait. Malathion sprays gave only a temporary reduction of *F. cannicularis*. (13 references.)
C. M. HARDWICK.

Chemical and biological examination of commercial pyrethrum extracts for insecticidal constituents. R. M. Sawicki and E. M. Thain (*J. Sci. Fd Agric.*, 1961, **12**, 137—145).—Three commercial pyrethrum extracts were examined chemically and biologically. The insecticidal constituents were extracted with nitromethane and separated by displacement chromatography. These fractions were identified chemically as cinerin I, pyrethrin I, cinerin II and pyrethrin II. "Reconstituted" pyrethrum solutions were biologically identical with solutions of commercial extracts containing the same quantities of pyrethrins and cinerins. The insecticidal activity of commercial pyrethrum extracts lies in the four known constituents pyrethrins I and II and cinerins I and II. (15 references.)
E. M. J.

Hercules AC-5727 as a residual spray for adult mosquitoes. J. H. Gahan, G. C. LaBrecque and H. G. Wilson (*J. econ. Ent.*, 1961, **54**, 63—67).—Deposits of *m*-isopropylphenyl methylcarbamate (1—10 mg./sq. ft.) gave 75—100% mortality in 24 h. 20 weeks after application. Increased dosages reduced exposure time so that a fresh deposit of 50 mg./sq. ft. produced immediate knockdown of *Anopheles quadrimaculatus* with 100-fold resistance to dieldrin. In the laboratory it was effective for longer periods than DDT, malathion, dieldrin or Bayer 29493 (*OO* dimethyl *O*-[4-(methylthio)-*m*-tolyl] phosphorothioate). Loss of effectiveness on whitewash and sun-dried brick was greater than that with DDT but similar to that with malathion. Hercules AC-5727 gave complete residual control of mosquitoes for 18 weeks in wooden animal houses.
C. M. HARDWICK.

Internal DDE production by normal and DDT-resistant larvae of *Aedes aegypti*. A. N. Chattoraj and A. W. A. Brown (*J. econ. Ent.*, 1960, **53**, 1049—1051).—The amount of DDE found in larvae of three DDT-susceptible larvae was $\frac{1}{10}$—$\frac{1}{4}$ as much as in three from DDT-resistant strains after exposure to 1 p.p.m. for 24 h. DDE production in one strain did not decrease when DDT-resistance was reduced by relaxation of selection pressure. (11 references.)
C. M. HARDWICK.

Colorimetric determination of halogenated nitrophenols added to streams as sea-lamprey larvicides. M. A. Smith, V. C. Applegate and B. G. H. Johnson (*Analyt. Chem.*, 1960, **32**, 1670—1675).—3-Trifluoromethyl-4-nitrophenol was determined by measuring its absorbance in alkaline solution at 395 mμ. A correction should be made for absorbance due to the stream-water. The max. error for 12 determinations in the range 2—6 p.p.m. was 0·2 p.p.m.
R. M. ROWLEY.

Microbial concentration of iron and manganese in low concentrations in water. R. S. Wolfe (*J. Amer. Wat. Wks Ass.*, 1960, **52**, 1335—1337).—The occurrence, physical structure and rough chemical analysis of a large micro-organism (believed to be *Clonothrix putealis*) containing very appreciable quantities of Fe and Mn are described. The organism flourishes in waters containing minute traces of Fe and Mn. B. F. FULLAN.

Treatment of combined sewage and fruit canning wastes. J. T. Norgaard, R. Hicks and D. A. Reinsch (*J. Wat. Pollut. Control Fed.*, 1960, **32**, 1088—1108).—Three basic processes were investigated: (*a*) anaerobic fermentation followed by activated sludge and high-rate filtration; (*b*) primary sedimentation followed by high-rate filtration; (*c*) activated sludge with and without sedimentation. The best method was (*c*), an activated sludge process with primary sedimentation and supplemented NH_3 feed during the peak periods.
C. A. SLATER.

Colour problems with beer waste. R. E. Pailthorp (*J. Wat. Pollut. Control Fed.*, 1960, **32**, 1201—1211).—High-rate trickling filters used in municipal sewage treatment plants do not remove enough colour from combined beer waste and domestic sewage to prevent its detection in a receiving stream. More beer colour is removed by increasing the recirculation ratio or decreasing filter loading. The latter increases B.O.D. Much Cl_2 is needed to remove beer colour, but the contact time could be short and no sludge would be produced. Lime treatment reduces the colour well but requires processing large vol. of sludge. O. M. WHITTON.

Culture of bacteria associated with effluent disposal. G. C. Ware (*Soc. chem. Ind.*, 1961, Monogr. 12, 165—174).—The bacteria involved in the biological treatment of sewage, and their mode of action, are discussed from the standpoint of their ease of culture. The possibility of their continuous homogeneous culture in sewage treatment is touched on. (35 references.) A. J. B.

Culture of *Oscillatoria* in organic wastes. A. C. Gaur, W. O. Pipes and H. B. Gotaas (*J. Wat. Pollut. Control Fed.*, 1960, **32**, 1060—1065).—A strain of *Oscillatoria* grew well in sewage and other org. wastes. Yields are similar to those of *Chlorella*, *Scenedesmus* and *Euglena*. The *Oscillatoria* removed 95—97% of the 5-day B.O.D. and 80—90% of the C.O.D.; the alga contained protein 40·9—44·4, ether-sol. fat 13·3—25·3 and ash 6·0—6·2%. It seems suitable for growth in waste-water stabilisation ponds. C. A. SLATER.

Effects of diverting the effluent from sewage treatment on the receiving stream. K. M. Mackenthun, L. A. Lueschow and C. D. McNabb (*Trans. Wis. Acad. Sci. Arts Lett.*, 1960, **49**, 51—72).—An effluent (~20 millions gal./day) was diverted into a small stream which had a flow of 9·6 cu. ft./sec. discharging into the river Yahara, thence into the Rock river. The flow of the stream was increased five-fold with substantial sludge deposits in some areas. Org. and inorg. N, P and B.O.D. increased; dissolved O_2 was reduced to a critical level. Phytoplankton populations were of substantially the same concn. between three stations of a given stream but greater in Rock than in Yahara and in the small stream were depressed. Submerged aquatic vegetation (filamentous algae) was affected. Bottom organisms indicated severe stream degradation, the population being reduced to tolerant sludge worms and midge larvae. The benthos showed a much greater response than the phytoplankton to the contents of the effluent. E. M. J.

Trend of caesium-137 in effluent of a large city. T. R. Folsom and G. J. Mohanrao (*Nature, Lond.*, 1960, **188**, 979—982).—Distribution of ^{137}Cs in raw and digested sewage was determined (Oct. 1959—June 1960) with a multi-channel γ-ray spectrometer at the plant (daily throughout 250 × 10^6 gal.). Total activity never reached a recognised tolerance level; concn. of input ^{137}Cs slowly decreased from ~1·4 to ~0·3 μμc./l., the half-purging time being ~17 days. Max batch-input concn. was 0·4 c.; enrichment factor after digestion was ~4·4 (dry basis). City's water-supply probably contributed ~6% of ^{137}Cs in sewage, food ingestion and garbage ~35%. The basic trend of ^{137}Cs is probably almost entirely dependent on fall-out Cs entering into food. There is 40-fold enrichment of Cs relative to K during digestion of sewage to dry fertiliser.
W. J. BAKER.

α-Substituted *OO*-dialkyl dithiophosphorylacetic esters. Montecatini Società Generale per l'Industria Mineraria e Chimica (B.P. 834,814, 6.6.56. It., 14.6.55).—Compounds $OR(OR')\cdot PS\cdot S\cdot CHR'''\cdot CO_2R''$ are claimed (R—R'' are saturated or unsaturated, straight- or branched-chain alkyl; R''' is aryl, e.g., naphthyl, optionally substituted). They have high insecticidal activity and are effective against rodents and parasites of plants and houses. The method of prep. of OO-*diethyl* S-(*carbethoxy*)-*phenylmethyl phosphorothiolothionate*, b.p. 149—150°/0·05 mm., is detailed. F. R. BASFORD.

Drying of sewage sludge. Dorr-Oliver Inc. (Inventor: M. W. Brandt) (B.P. 835,216, 7.3.55).—Digested sewage sludge is conditioned for pelleting (for use as saleable fertiliser) by reducing the water content to 45—55 wt.-%, then mixing with dry material (e.g., drier sludge), and stirring. The resulting pellets (optionally enriched with additional fertiliser) are finally dried in fluidised bed to give a non-dusty product of ≯5 wt.-% of water.
F. R. BASFORD.

4.—APPARATUS AND UNCLASSIFIED

Determination of traces of copper in rubber latex ash by neutron activation analysis. J. A. W. Dalziel and R. C. H. Hsia (*J. Sci. Fd Agric.*, 1961, **12**, 127—130).—The radiochemical purification needed for the β- and γ-assay of ^{64}Cu was examined. A single sulphide pptn. from latex ash does not give a pure source of ^{64}Cu, but γ-counting was effected by measurement of the decay of the 0·51 MeV photo peak by scintillation spectrometry. β-counting was effected after further purification of the ^{64}Cu from ^{32}P by electrodeposition. The chemical yields of Cu in the counted sources were determined by titration with EDTA. Out of 10 results 7 gave a mean value of 370 ± 20 p.p.m. of Cu in the ash. E. M. J.

Journal of Applied Chemistry

The following papers are appearing in the June, 1961, issue

Correlation between surface tension and other physical properties
By Franklin J. Wright

Studies in hydrogen-bond formation. IX. Bonding between esters and the chloro-group
By C. H. Giles and S. N. Nakhwa

Reactivity of lime and related oxides. VII. Crystal size variations in calcium oxide produced from limestone
By D. R. Glasson

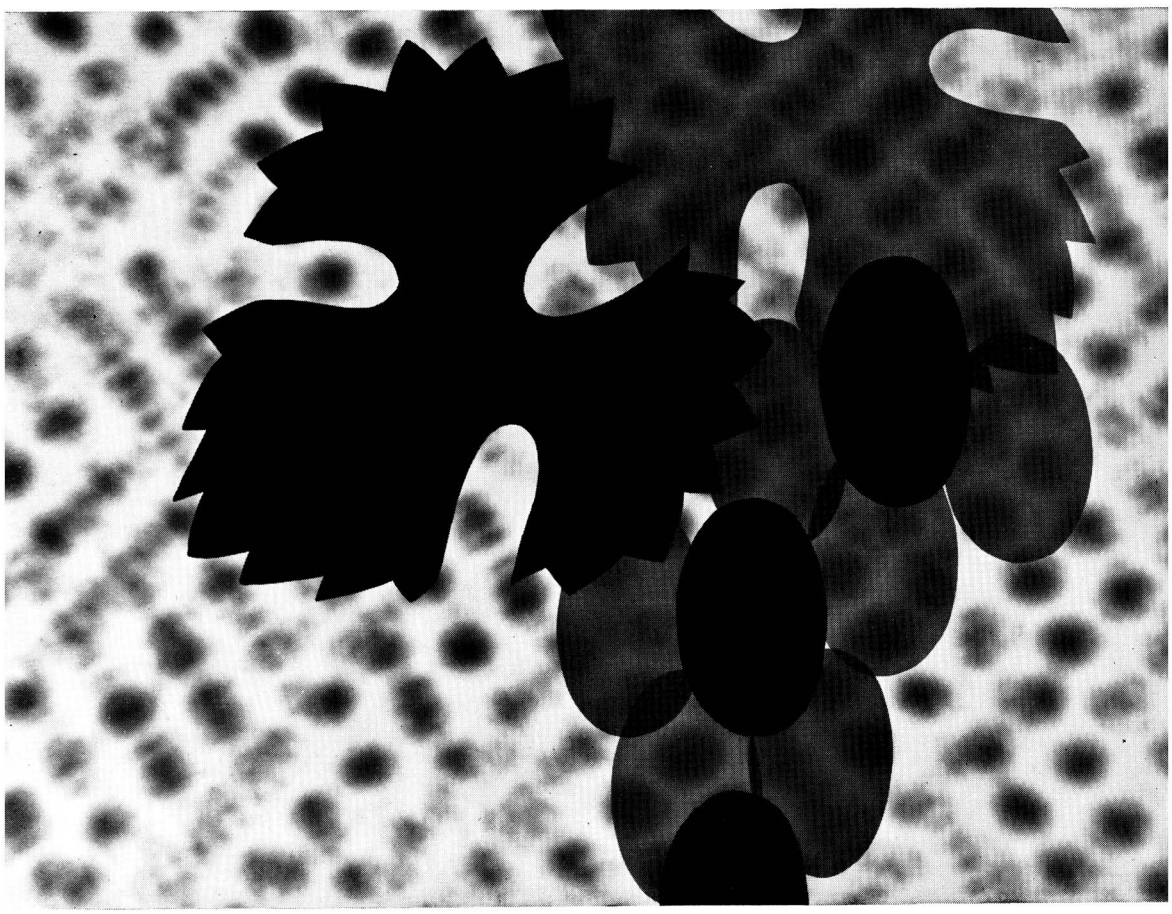
The transformation of bayerite into hydrargillite
By Taichi Sato

The pyrolysis of cellulose and the action of flame-retardants
By F. H. Holmes and C. J. G. Shaw

The removal of copper extrusion sheaths from zirconium and its alloys
By I. A. Menzies and F. Rigby

Vapour–liquid equilibria in the system water–propionic acid
By P. Dakshinamurty, G. Jayarama Rao and C. Venkata Rao

Grape (grēp): prob. f. OF. *graper* to gather with a vine hook, f. *grape* hook. Genus *Vitis* of family *Vitaceae*, of which oldest, most cultivated species is *V. vinifera*.1. Prob. derived f. Caspian Sea area, thence to Asia Minor, Greece, Sicily. Intro. to France by Phoenicians *circa* 600 B.C. 2. Seriously attacked by many pests including insects and nematodes. Needs protection up to harvest. 3. During critical close-to-harvest period Phosdrin is the *only* insecticide usable without harmful residues. 4. Against pests of vine Shell pesticides Phosdrin, aldrin, dieldrin and Nemagon offer greatest protection. **Phosdrin** *Trade Mark*



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