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Bigger, better yams for sale

To Onitsha market in Eastern Nigeria comes the produce of the world . . . bicycles from Birmingham, watches from Switzerland, sewing machines from Scotland, typewriters from Italy . . . yams from Nigeria itself. Yams that are plentiful. Yams the beetles didn't get . . .

Yams are important to Nigeria as a reserve food. They are high yielding and store well after harvest. If they get to harvest, that is . . . for yams are also important to *Heteroligus meles*, chief of the beetle pests that prey upon them. These beetles are a serious menace and, since they attack the tuber beneath the soil surface, difficult to eradicate.

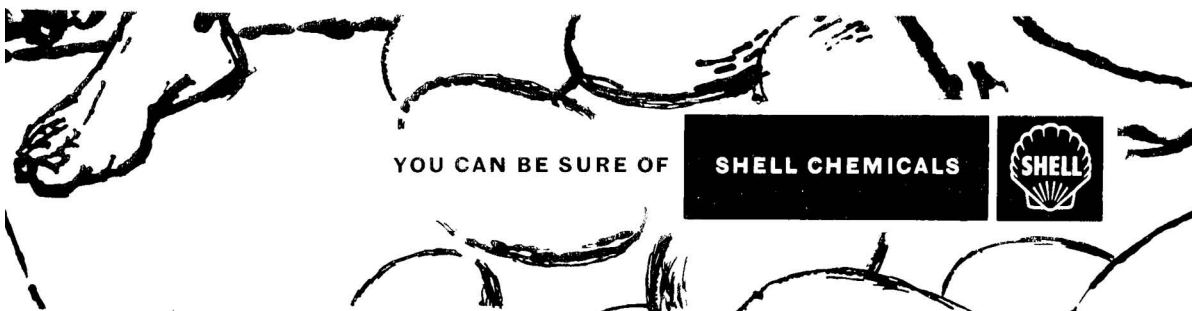
But tests have shown that, with aldrin, yam cultivation can be put on a sound footing. Of all insecticides used, aldrin gave the

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EFFECTS OF NITROGEN AND POTASSIUM FERTILISERS ON THE MINERAL STATUS OF PERENNIAL RYEGRASS (*LOLIUM PERENNE*). II.*—Anion-Cation Relationships

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

A number of anion-cation relationships including AA, TA, VA, EA (defined in the text), $K/(Ca + Mg)$ and $(K + Ca)/Mg$ were calculated from the results of a previous paper.⁷ In addition a number of samples obtained from 'normal' and 'tetany' areas were examined. The results indicated that AA, TA and VA values and the $K/(Ca + Mg)$ ratio were increased by both nitrogen and potassium fertiliser treatments. In a number of samples from 'tetany' areas the $K/(Ca + Mg)$ ratios did not approach 2.2, although the AA values were generally high.

Introduction

Brouwer and co-workers¹ have discussed the importance of considering the acid-base equilibrium in herbage in relation to certain metabolic disturbances which occur in ruminant animals. Among the relationships considered (expressed in mequiv./kg. of dry matter) the following have been presented:

Alkali alkalinity (AA) = Na + K - Cl - S

Alkaline earth alkalinity (EA) = Ca + Mg - P

Total alkalinity (TA) = (Na + K - Cl - S) + (Ca + Mg - P)

Base excess (VA) = (Na + K - Cl - S) - (Ca + Mg - P)

In addition to the above ratio $K/(Ca + Mg)$ has been calculated. This ratio was employed by Verdeyen² who observed that grass tetany in dairy cattle was particularly liable to occur when the ratio in the pasture rose above 2.20. Kemp & t'Hart³ have also confirmed this. Although the above ratio has been used by a number of workers, it seems logical to consider the antagonism of potassium and calcium to magnesium. With this in view the ratio $(K + Ca)/Mg$ has also been considered.

Brouwer^{1a} has studied the anion-cation relationships for a number of grass and hay samples and from the values obtained he has suggested the following tentative limits for normal grass and hay:

AA 100-375; EA 50-325; TA 300-550; VA - 150 to +250

The grasses obtained from tetany-inducing pastures were characterised by negative EA values and extremely high AA and VA values, the TA values being intermediate ones. Brouwer & van de Vliert^b have shown in one series of investigations that the VA value from tetany pastures may be very high and quoted values up to +600 mequiv./kg. of dry matter. It is interesting to compare this value with those from haemoglobinurea pastures inducing acid urine reaction in which the VA was negative (to -600). Brouwer^{1a} has also stated that potassium fertilisers were mainly responsible for the fluctuation of the AA and EA values. Bosch⁴ has stated that 'excess of a base' may be lowered by some fertilisers, so that diseases (e.g., scouring) are more likely to occur.

More recently Brandsma⁵ has studied the mineral composition of grass on 14 'normal', healthy dairy farms. From a total of 150 samples he calculated the following:

AA 145 ± 109; EA 118 ± 80; TA 363 ± 128; VA 128 ± 142

The mean K, Ca and Mg values in the herbage dry matter from these farms were 2.97, 0.69 and 0.22%, respectively. The $K/(Ca + Mg)$ ratio (in mequiv./kg. of dry matter) calculated from these figures is 1.44.

All these anion-cation relationships have been calculated from data collected from investigations in the Netherlands. There is little indication as to how these 'normal' values can be applied to farming conditions in Scotland, since as far as the authors are aware no similar

* Part I: *J. Sci. Fd Agric.*, 1960, **11**, 422

surveys have yet been completed. The main purpose of this paper is to consider some anion-cation relationships in perennial ryegrass grown under different nitrogen and potassium fertiliser treatments. The percentage composition of these ryegrass samples has been reported in a previous paper.⁶ Throughout these calculations, one g.-atom of P has been valued at 3 in agreement with Brouwer's suggestion.^{1a}

Experimental

The design of the experiment, sampling and analytical techniques have been described in Part I⁶ and the composition of the perennial ryegrass samples has been calculated in mequiv./kg. of dry matter from the data there reported. In addition to the above, a number of samples were collected from different areas in S.E. Scotland. Nine samples of herbage were taken from apparently 'normal' farms in which no previous history of hypomagnesaemic tetany had been recorded. Eight samples of herbage were taken from five different farms on which deaths from tetany had occurred, while animals had been grazing. The samples were collected within a few days of the onset of tetany symptoms.

Results

The results are shown in Table I and the anion-cation relationships calculated from these results in Table II.

Table I

Composition of perennial ryegrass, mequiv./kg. of dry matter

Treatment*	1st cut (18.7.56)					2nd cut (20.8.56)					3rd cut (23.5.57)										
	Ca	Mg	Na	K	Cl	P	S	Ca	Mg	Na	K	Cl	P	S	Ca	Mg	Na	K	Cl	P	S
O	275	192	35	860	116	387	269	235	167	22	873	130	436	325	210	100	22	581	161	329	194
K	250	125	35	768	217	397	175	230	167	22	888	164	458	256	210	100	26	609	189	310	194
C	280	158	35	860	147	290	219	295	192	30	1075	135	465	269	235	100	26	632	158	339	150
KC	235	125	57	968	116	358	194	245	183	22	993	175	410	275	200	100	22	627	166	300	188
N ₁	261	142	57	606	104	290	181	275	175	61	1239	107	416	244	260	125	74	625	138	368	194
N ₁ C	235	150	57	765	124	348	163	265	183	57	1118	135	513	288	240	117	48	686	141	368	194
N ₁ K	230	133	57	765	124	358	213	225	158	30	1021	152	416	238	190	117	22	765	155	358	194
N ₁ KC	235	142	35	827	116	310	181	250	200	26	1224	127	436	250	210	117	22	778	166	378	194
N ₂	235	142	74	829	99	319	256	270	183	83	1129	96	494	250	260	133	91	653	152	397	206
N ₂ C	260	150	117	765	113	348	188	255	192	96	1070	121	474	238	270	142	104	748	133	407	213
N ₂ K	230	150	87	963	135	339	188	270	175	65	1265	104	445	256	220	133	30	876	138	387	206
N ₂ KC	220	133	61	896	149	348	213	220	175	57	1354	127	513	269	210	133	26	934	144	387	219

Table II

Cation-anion relationships in perennial ryegrass calculated from mequiv./kg. of dry matter

Treatment*	1st cut				2nd cut				3rd cut				
	AA	EA	TA	VA	AA	EA	TA	VA	AA	EA	TA	VA	
O	516	80	590	430	1.84	5.91	440	-34	406	474	2.17	6.63	248
K	411	-22	380	433	2.05	8.14	490	-61	429	551	2.24	6.69	252
C	536	148	678	382	1.96	7.22	566	22	588	544	2.21	7.14	350
KC	715	2	717	713	2.69	9.62	565	12	577	553	2.32	6.77	295
N ₁	468	113	581	355	1.73	6.74	949	34	983	915	2.75	8.65	367
N ₁ C	455	57	512	398	1.89	6.80	752	-65	687	817	2.50	7.56	399
N ₁ K	485	5	490	480	2.11	7.48	661	-33	628	694	2.67	7.89	436
N ₁ KC	565	67	632	498	2.19	7.48	873	14	887	859	2.72	7.37	440
N ₂	548	58	606	490	2.20	7.49	866	-41	825	907	2.49	7.64	386
N ₂ C	581	62	643	519	1.87	6.83	807	-27	780	834	2.36	6.90	506
N ₂ K	727	51	778	676	2.53	7.95	970	0	970	970	2.84	8.77	562
N ₂ KC	595	5	600	590	2.54	8.39	1015	-118	897	1133	3.43	8.99	597

* for explanation of symbols see Part I (preceding paper)

The AA, TA and VA figures generally were increased by both nitrogen and potassium treatments. The highest values were obtained in samples from the plots which had received heavy dressings of nitrogenous fertiliser as well as potassium. The AA, TA and VA values for the second cut (August, 1956) were very high compared with those for the other two cuts.

Negative EA values were obtained in samples from most treatments at the second and third cut. The lowest values in these cuts occurred in the grass which had received both nitrogen and potassium. In the first cut only one negative EA value occurred and while the effect of nitrogen was irregular, potassium reduced the EA values consistently.

Application of potassium sulphate consistently increased the K/(Ca + Mg) ratio at all cuts. Some very high ratios were obtained in the second cut, particularly with potassium combined with high nitrogen treatments. The highest value (3.34) was obtained from the N₂KC plots. The (Ca + K)/Mg ratios were not consistently affected by different treatments.

The results of the farm herbage samples and details of the species are given in Table III. Samples 9-13 were taken from hill land which had not received any fertilisers.

Table III

Mineral composition and cation-anion relationships in some miscellaneous herbage samples

Samples from 'tetany' areas No.	Herbage	mequiv./kg. of dry matter										Cation-anion relationships		
		Ca	Mg	Na	K	Cl	P	S	AA	EA	TA	VA	K/(Ca+Mg)	(K+Ca)/Mg
1	Ley (mixed spp.)	240	133	35	202	110	242	156	-29	131	102	-160	0.54	3.32
2	Ley (" ")	280	183	187	755	107	368	194	641	95	736	546	1.63	5.66
3	Ley (" ")	300	142	35	535	93	310	156	321	132	453	189	1.21	5.88
4	Hill P.P.	285	167	30	310	158	397	219	-37	55	18	-92	0.69	3.56
5	Ley (" ")	275	183	91	781	113	484	231	528	-26	502	554	1.71	5.77
6	Ley (mixed spp.)	335	142	61	904	127	426	156	682	51	733	631	1.90	8.73
7	Ley (" ")	335	158	39	614	110	465	206	337	28	365	309	1.25	6.01
8	Ley (" ")	360	175	326	783	82	532	263	764	3	767	761	1.46	6.53
* Normal ' samples														
9	<i>Molinia caerulea</i>	120	158	13	456	104	186	206	159	92	251	67	1.64	3.65
10	<i>Nardus stricta</i>	90	83	17	253	79	174	88	103	-1	102	104	1.46	4.13
11	<i>Festuca ovina</i>	105	125	30	402	79	252	125	228	-22	206	250	1.75	4.06
12	Hill P.P., mainly <i>Molinia</i> , <i>Nardus</i> ,	55	67	22	192	37	271	94	83	-149	-66	232	1.57	3.69
13	<i>Festuca</i> spp.	80	100	13	430	96	203	106	241	-23	218	264	1.57	5.30
14	Ley (mixed spp.)	230	150	30	397	93	319	156	178	61	239	167	1.04	4.18
15	Ley (" ")	275	183	35	532	99	436	294	174	22	196	152	1.16	4.41
16*	P.P. (mixed sp., well fertilised)	280	142	52	919	79	397	225	667	25	662	642	2.18	8.44
17	P.P. (" " , mainly <i>Festuca rubra</i>)	305	183	244	463	20	319	188	499	169	668	330	0.95	4.20

* cases of tetany reported the following season

Discussion

It has previously been mentioned that the 'normal' farms had no previous record of hypomagnesaemic tetany. The field from which sample 16 had been obtained had received heavy dressings of fertilisers and it was reported in the year following sampling that three deaths had occurred in a herd of cows while grazing this particular field. In spite of the fact that no tetany had been recorded at the time of sampling it was obvious from the subsequent events that this sample was not 'normal'. The abnormality of this sample is shown in the high AA, TA and VA values as compared with Brouwer's normal range. The K/(Ca + Mg) ratio is relatively high and approaches Verdeyen's critical level of 2.20.

With the exception of the above sample most of the AA and VA values fall within Brouwer's normal limits although the TA values with one exception are abnormally low. Amongst the 'normal' samples, four show negative values for EA. These all occur in hill species (*Nardus* and *Festuca* spp.).

With the exception of sample 16 referred to above, all the (K + Ca)/Mg values are below 5.2 whereas most of the samples from 'tetany' areas show values above this level. On the other hand, the K/(Ca + Mg) ratios are generally lower in the 'tetany' than in the normal samples. No values approaching 2.2 occur in the 'tetany samples' and it is clear that this ratio is no indicator in the small number of samples examined.

Many of the grasses obtained from tetany-inducing pastures were characterised by high AA values in agreement with Brouwer's observations. The TA and VA values showed similar trends to the AA figures although few were abnormally high according to Brouwer's limits. Only one negative EA value occurred in grass from the 'tetany' areas.

It is obvious that since only a few farm samples have been examined it is impossible to draw any definite conclusions from the results and it is clear that a much wider survey of tetany and normal areas in this country is necessary.

Table II shows the anion-cation relationships for the experimental samples of perennial ryegrass. The most outstanding features of these results are the exceptionally high AA, TA and VA values which occurred in most of the samples. It is difficult to explain why many of these values should be high in the control and the non-fertilised ryegrass-clover plots when compared with Brouwer's¹⁶ and Brandsma's⁵ normal values. Possibly the fact that the present analyses were confined to a single species of grass, whilst the Dutch workers' normal values were

obtained from mixed species, may be an important factor. There are few data available for the mineral composition of different species of grasses although it is clear from the work of Thomas & Thompson⁷ that considerable variations between species can occur.

The majority of the EA values fall below Brouwer's lower limits for normal herbage and in the second cut all these values were negative which, according to Brouwer,^{1a} is characteristic of tetany-inducing pastures. The high nitrogen-potassium treatments consistently produced the highest AA, TA and VA values at all cuts. These values were particularly high at the second cut. It is interesting to note that these high values are not necessarily associated with low herbage-magnesium content since the second cut gave samples with considerably higher magnesium content than the other two cuts (Table I). Although the high nitrogen-potassium treatments generally gave low EA values, the results were not as consistent as the other values. Table III shows that the EA values from 'tetany' and 'normal' areas are erratic. The calculation of EA does not take potassium directly into account and this may be responsible for the apparent lack of significance of this value.

The majority of the $K/(Ca + Mg)$ ratios are above Verdeyen's limit² of 2.2. According to this worker and Kemp & t'Hart³ these samples of herbage would tend to induce tetany in stock. All the values in the second cut, except the control, were above 2.2. The highest levels at all cuts were obtained on the herbage from the high nitrogen-potassium plots.

The $(K + Ca)/Mg$ ratios in the farm samples from tetany areas (Table III) were generally higher than in the normal samples and in fact this ratio appears to be a better guide than the $K/(Ca + Mg)$ ratio. In the experimental samples the $(K + Ca)/Mg$ ratios were all high and were generally increased by potassium fertiliser.

Conclusions

From a consideration of a number of anion-cation relationships calculated from the results of the previous paper on perennial ryegrass it has been shown that

(1) AA, TA and VA values were increased by both nitrogen and potassium fertiliser treatments. A large number of negative EA values were obtained.

(2) The $K/(Ca + Mg)$ ratio was consistently increased by applications of potassium and nitrogen.

(3) In a number of herbage samples taken from 'tetany' and 'normal' farms, none of the $K/(Ca + Mg)$ ratios from the 'tetany' samples approached 2.2. The $(K + Ca)/Mg$ ratios, however, generally gave a better indication of tetany-inducing herbage.

(4) Most of the samples obtained from tetany areas were characterised by high AA values. The TA, VA and especially EA values were less conclusive.

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PILOT-PLANT STUDIES ON THE PREPARATION OF CRUDE PAPAIN FROM RAW PAPAYA

By G. V. KRISHNAMURTHY, B. S. BHATIA, GIRDHARI LAL and V. SUBRAHMANYAN

The results are presented of an investigation of the yield and total solids content of the latex obtained in successive tappings of papaya fruit and of the yield and activity of the crude papain obtained from the latex samples by sun- and vacuum-drying. The effect of the addition of metabisulphite, with and without thymol, on the activity of the dried products is described.

Introduction

Studies on the preparation of papain have been conducted by several workers,¹⁻⁴ but little information is available in the literature regarding the yield and total solids content of the latex and the yield and activity of crude papain in different individual tappings, except for papers from workers in Ceylon.⁵ These points have now been investigated, with a study of drying conditions and chemical treatment of papaya latex in a pilot-plant investigation on the development of an integrated process for the preparation of crude papain and pectin* from raw papaya.

Experimental

From the third-year crop of a papaya plantation at the Sewage Farm, Mysore, raw papaya fruits at a stage of maturity of about 3 months after setting were selected, divided into 20 batches and marked for tapping. Fruits in each batch were obtained from four to six separate trees. Data regarding number of fruits, weight of fruits and average weight of fruit in different batches are given in Table I. Tapping was done during January and February 1959. Fruits in each batch were lanced with stainless steel knives to give four to six longitudinal cuts about $\frac{1}{2}$ in. deep. The latex was collected in stainless steel shallow plates. At the end of each tapping, the latex which coagulated on the surface of the fruit was scraped and added to the batch. Fruits in each batch were tapped six times in the course of 16 days, i.e., 1st, 3rd, 6th, 9th, 12th and 16th day after the start, except that for the first five batches only four tappings were done. Since the last 15 batches were each tapped six times and were comparable with each other, the conclusions are based on 15 batches only.

Table I

Number and average weight of fruit in 15 different batches

	Range	Average
Number of fruits in each batch	66-128	97
Total weight of fruits in each batch, lb.	154-364	277
Average weight of fruit in each batch, lb.	1.6-5.5	3.0

Latex from each tapping was weighed, passed through a 50-mesh sieve, mixed with 0.5% of potassium metabisulphite (KMS), or 0.5% of KMS + 0.2% of thymol where necessary, and dried on aluminium trays, either in the sun (temperature 25-40°, R.H. 30-75% and drying time 5-6 h.) or in a vacuum shelf dryer at 55° and 28 in. of vacuum (drying time 3-4 h.). The tray loading in each case was about 3 oz. of latex per square foot of drying area. The total solids in the latex were calculated from the loss in weight during vacuum drying. The dried samples of crude papain which contained about 5% of moisture (loss on drying at 70° for 6 h.), were packed in glass jars with metal screw lids, waxed and stored at 0° F until required for analysis. Papain activity was determined by milk clotting by the Blau modification⁶ of the method of Balls & Hoover⁷ with skim milk powder as the substrate.

* Work done on pectin is being published elsewhere

Results and discussion

The range and average values for yield of latex and crude papain and total solids in latex of six separate tappings in 15 batches, and values for milk clotting activity of the crude papain based on 8 batches, are given in Table II.

Table II

Results for separate tappings in 15 different batches*

Tapping No.	Yield of latex, % of fruit		Yield of crude papain, % of fruit		Total solids % in latex		Milk clotting units of native activity per g. of crude papain		
	Range	Average	Range	Average	Range	Average	Range	Average	
1	0.15-0.51	0.27	0.030-0.120	0.061	11.1-29.8	23.1	250-400	294	
2	0.10-0.36	0.18	0.036-0.160	0.073	32.1-54.9	40.8	250-364	302	
3	0.07-0.27	0.16	0.017-0.066	0.044	15.2-40.5	27.6	190-364	272	
4	0.06-0.18	0.12	0.016-0.064	0.038	14.7-50.0	30.6	190-286	242	
5	0.04-0.14	0.09	0.017-0.048	0.029	15.5-44.3	33.1	166-286	232	
6	0.04-0.22	0.11	0.015-0.044	0.031	17.9-47.6	31.0	190-334	252	
Total of 6 tap-pings, i.e., in one batch		0.54-1.57	0.94	0.15-0.47	0.28	26.1-34.2	29.5	—	—

* Eight batches for milk clotting activity of crude papain. In all these batches 0.5% potassium metabisulphite was added to the latex before drying in a vacuum shelf dryer

(1) Yield of latex

The average yield of latex was higher at the 0.1% level of significance in tapping 1 than in tappings 2 to 6. Values for tappings 2 and 3 were significantly higher than for tappings 4-6 (0.1% level). Balls *et al.*⁴ reported a range of 0.3-1.0% yield of fresh latex based on lancing of six individual fruits. In the present experiments a range of 0.04-0.51% in six separate tappings from the lancing of 66-128 fruits (average 97 fruits) was obtained. The average yield of latex per batch was 0.94% (range 0.54-1.57%).

(2) Yield of crude papain

The average yield from the second tapping (0.073%), although higher than that from the first tapping (0.061%), was not statistically significant. Yields from the first and second tappings were higher than yields from the third and fourth tappings at the 0.1% level of significance, while the yields from the latter were significantly higher than those from the fifth and sixth tappings (5% level). From this it is concluded that the yield is the highest in the first two tappings and gradually decreases in subsequent tappings. A range of 0.07-0.12% yield of crude papain based on lancing of six separate fruits has been reported.⁴ The corresponding range in the present experiments is 0.015-0.160 based on lancing of 66-128 fruits (average 97) at a time. The average batch yield from six tappings is 0.28% (range 0.15-0.47%). The batch yield for six tappings was about 25% higher than the batch yield from four tappings. It is obviously desirable to tap the fruits six times.

(3) Total solids in latex

The yield of crude papain is dependent on the yield of latex and its total solids content. Although the yield of latex is less in tapping 2 than in tapping 1, because of the higher total solids content of tapping 2, its yield of crude papain is higher than that of tapping 1. In the literature, the total solids content of latex has been reported to be 14.9-18.4%.^{1, 4} In the present experiments the range is much wider, i.e., 11.1 to as high as 54.9% in separate tappings.

The total solids content for the second tapping is significantly higher, and that in the first tapping significantly lower (both at 0.1% level), than in any other tapping. The highest value for the total solids content in the second tapping may probably be explained by the sudden increase in the physiological activity of the fruits resulting from the lancing injury at the first tapping. After the second tapping the physiological activity probably settles down to a level which is lower than that at the second tapping, but higher than that in the first tapping.

(4) *Activity of crude papain*

Statistical examination of the data regarding the activity of the crude papain showed that values for the first and second tappings were similar and they were significantly higher than the others (0.1% level). There was little increase in the activity after activation.

(5) *Effect of drying conditions and the chemical treatment of latex on the activity of crude papain*

It is clear from Table III that addition of 0.5% metabisulphite (KMS) to the latex before sun-drying helps considerably in increasing the activity of crude papain. In vacuum drying, however, only slight improvement in the activity is obtained by the addition to the latex of 0.5% KMS with or without 0.2% of thymol. In Table IV, the effect of treatment of the latex with (i) 0.5% KMS and (ii) 0.5% KMS + 0.2% thymol on the activity of crude papain in four separate tappings is compared. Statistical examination of the data showed that in all the tappings, there was no significant difference between the two treatments. Hinkel² found treatment (ii) to be the best for high retention of the activity of crude papain. He, however, did not compare this treatment with the use of 0.5% KMS without thymol, but only with a lower concentration (0.1%) of KMS. Thus it appears that addition of 0.2% thymol to the latex treated with 0.5% KMS does not impart any additional advantage during the drying stage.

Table III

Effect of drying conditions and the chemical treatment of papaya latex on the activity of crude papain

No.	Drying method	Chemical treatment	Milk clotting units ⁶ of native activity per g. of crude papain
1.	Sun-dried	Nil	111
2.	" "	0.5% KMS*	200
3.	Vacuum dried	Nil	222
4.	" "	0.5% KMS	250
5.	" "	0.5% KMS + 0.2% thymol	286

* KMS = potassium metabisulphite

Table IV

Comparison of treating the latex with 0.5% metabisulphite with and without 0.2% thymol on the activity of the crude papain in four separate tappings

Tapping No.	Milk clotting units of native activity per g. of crude papain							
	Latex treated with 0.5% KMS* and vacuum dried				Latex treated with 0.5% KMS + 0.2% thymol and vacuum dried			
	1	2	3	4	1	2	3	4
Range	250-400	250-400	190-364	190-286	308-500	250-444	222-308	210-250
Average†	300	312	276	250	396	345	254	226

* KMS = potassium metabisulphite

† Average of 10 batches in case of latex treated with 0.5% KMS and 4 batches in latex treated with 0.5% KMS + 0.2% thymol

Conclusions

The results of pilot-plant studies on the preparation of crude papain, yield and total solids in latex and yield and activity of crude papain in six separate tappings of 20 batches of raw papaya, show that

(i) the yield of latex is the highest in tapping 1 and least in tappings 4-6. The yield per batch varies from 0.54 to 1.57% with an average of 0.94% ;

(ii) the yield of crude papain is the highest in the first two tappings and gradually decreases in subsequent tappings. The yield per batch varies from 0.15 to 0.47% with an average of 0.28% ;

(iii) the total solids content of the latex is the highest in the second tapping and the lowest in the first. Total solids vary from 11.1 to 54.9% ;

(iv) the milk clotting activity of the crude papain is highest in the first two tappings.

Addition of 0.5% metabisulphite to the latex improves considerably the activity of the crude papain when prepared by sun-drying and only slightly when prepared by vacuum drying. No further improvement in the activity of crude papain is obtained by adding 0.2% thymol to the latex in addition to 0.5% metabisulphite.

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BONE-TAINT IN BEEF.—II.* Bacteria in Ischiatic Lymph Nodes

By P. M. NOTTINGHAM

In a survey of the bacteria in ischiatic lymph nodes taken from freshly-killed cattle, a number of types of both Gram-positive and Gram-negative organisms were isolated. The average number of aerobic bacteria per lymph node appears to be related to the mean rainfall for the two months preceding slaughter.

Two outbreaks of bone-taint involving six sides of beef were studied. Potential taint-producing bacteria isolated from the tainted meat included bacilli, coliforms, pseudomonads and clostridia. In the second outbreak the spoilage was mainly due to anaerobes.

The bacterial flora of lymph nodes and tainted meat were found to have much in common. This is considered to be further evidence for the view that the infection which causes bone-taint may be present in the lymph nodes during life, and spread to the surrounding tissues after death.

Introduction

Bone-taint, or the deep-seated spoilage of meat, has been shown to be of bacterial origin. Previous workers have found that, although more than one organism may be responsible for the development of the putrid or sour odours characteristic of bone-taint, the bacteria most frequently implicated have been members of the family *Bacillaceae*.¹⁻⁴ It has been suggested that the spoilage bacteria enter the deep tissues from the gut during life, or at death through infection of the blood-stream by the sticking knife or other means.^{5, 6} More recently, evidence

* Part I: *J. Sci. Fd Agric.*, 1956, **7**, 546

has been put forward to support the view that the infection producing bone-taint is present in the lymph nodes at the time of slaughter.^{7, 8} In a previous communication from this laboratory, Cosnett *et al.*⁸ have shown that a considerable proportion of ischiatic lymph nodes taken from cattle carcasses immediately after slaughter contain Gram-positive organisms similar to those found in tainted meat, and they observed a correlation between the incidence of Gram-positive rods in these lymph nodes and the rainfall during the two months prior to slaughter.

In the present work, a more detailed study of the bacteria in ischiatic lymph nodes from cattle has been made. Isolations from lymph nodes and tainted meat have been compared.

Experimental

Preparation and examination of lymph nodes

The ischiatic lymph node, which lies in fatty tissue on the outer aspect of the sacro-sciatic ligament, can be removed without difficulty following the longitudinal splitting of the carcass. This node, embedded in fat, was removed about 30 min. after slaughter and placed in a sterile container for transport to the laboratory. Only one node was taken from each carcass, and any that were cut or damaged during removal were rejected. About one hour post-mortem, the whole fat-encased node was dipped in 95% ethanol and held over a flame until the alcohol had burnt off. This procedure was repeated, then the fat was removed under aseptic conditions. The exposed surface of the node was then seared in a similar manner. The node was cut into small pieces with sterile scissors, before being homogenised with 50 ml. of sterile 0.85% saline for 2 min. in the small metal cup of a Waring Blender.

Four nutrient agar plates were poured with 1-ml. portions of the homogenate. Two of these were incubated at 37°, and two at 25°, for 3 and 5 days respectively, before being counted and a number of colonies were selected and subcultured for further examination. About 5 ml. of the homogenate were added to cooked meat medium (Difco) and incubated at 37° for 2-3 days. Smears for microscopical examination were made from these cultures. Tubes of liver veal agar (Difco), and liver veal agar containing 0.03% sodium azide, were inoculated from the cooked meat cultures, and heated at 80° for 10 min., before incubation at 37° for 2-3 days. Colonies of aerobic and anaerobic spore-forming organisms were picked from tubes showing growth.

Lymph nodes were obtained from a total of 162 beef carcasses and smears for microscopical examination were made from all these samples. Aerobic counts were made on 134 nodes and colonies were picked for further examination from 42 nodes.

Tainted carcasses

Samples of meat (2-3 lb.) were cut from the tainted areas and removed to the laboratory, where they were trimmed and seared, before several 5-g. samples were taken aseptically from the interior. The popliteal lymph node was obtained from three of the tainted sides. Direct smears, enrichment cultures in cooked meat medium and nutrient agar plate counts were carried out on these samples.

Results

Ischiatic lymph nodes

Microscopical examination of smears from plate and enrichment cultures of 162 ischiatic lymph nodes showed that 93 (57%) contained Gram-positive cocci, 84 (52%) contained Gram-positive rods and 34 (21%) Gram-negative rods. Bacilli were found in 21 (13%) and clostridia in 11 (6.8%). Only 18 (11%) appeared to be sterile. It was not possible to isolate and characterise more than a small proportion of these organisms and, in general, attention was concentrated on the spore-forming bacteria.

A total of 94 organisms was picked out for further examination from plate and liver veal agar tube cultures from 42 nodes. Of these, 11 were not identified and 16 were discarded as duplicates. The remaining 67 different organisms were identified as far as possible in accordance with Bergey's scheme of classification.⁹ The seven genera that were present, together with the number of isolates of each genus and the number of samples from which each was isolated are given in Table I.

Of the aerobic spore-forming bacteria, *Bacillus licheniformis* and *Bacillus cereus* were the most common types, the former being isolated from five nodes and the latter from four. *B. pumilus* and *B. subtilis* were found in three nodes, *B. coagulans* in two and *B. macerans* in one. Two nodes contained at least two species of bacillus.

Table I

Distribution of genera isolated from ischiatic lymph nodes and tainted beef samples

Genus	Number of isolates		Number of samples from which each genus was isolated						
	Lymph nodes (42)	Tainted beef samples (21)	Lymph nodes (42)	1 (4)	2 (5)	3 (4)	4 (4)	5 (2)	6 (2)
<i>Escherichia</i>	2	9	2	1	2	2	—	1	2
<i>Aerobacter</i>	3	1	3	1	—	—	—	—	—
<i>Proteus</i>	—	1	—	—	—	—	1	—	—
<i>Achromobacter</i>	7	4	6	—	2	1	—	—	—
<i>Micrococcus</i>	30	5	17	1	—	1	—	1	2
<i>Pseudomonas</i>	5	4	4	3	—	1	—	—	—
<i>Bacillus</i>	18	8	16	—	2	1	1	—	—
<i>Clostridium</i>	3	27	3	4	3	3	4	2	1

The figures in parenthesis indicate the number of samples examined in each case

The average counts of aerobic bacteria present in the ischiatic lymph nodes collected each month in the period February to August, 1957, together with the percentage of nodes containing Gram-positive rods, are given in Table II. The mean rainfall of the two preceding months for the observation points Waingawa and Palmerston North, representative of the farming districts of Wairarapa and Manawatu, the source of almost all the cattle, is also shown in this Table.

Table II

Mean monthly counts of bacteria in ischiatic lymph nodes

Month	No. of nodes examined	Log ₁₀ numbers/node		% of nodes containing Gram-positive rods	Mean rainfall (in.) during two preceding months
		at 37°	at 25°		
February	7	2.93	2.65	57	4.0
March	12	3.54	3.28	42	1.7
April	19	2.93	3.20	58	2.4
May	24	2.60	3.11	58	3.2
June	24	2.40	2.78	50	4.2
July	28	2.40	2.74	29	4.7
August	20	2.60	3.08	85	3.9

No correlation was observed between the percentage of nodes containing Gram-positive rods and the rainfall for the one or two months preceding the month of slaughter. However, when the logarithm of the average count at each temperature for each month was compared with the mean rainfall for the two preceding months a significant correlation was apparent. After a period of low rainfall the numbers of bacteria present in the ischiatic lymph nodes were higher than when the rainfall for the two months prior to slaughter had been high. The 37° count appeared to be related to the rainfall of both the preceding months and the mean of the two preceding months. In both cases $r = -0.84$, equivalent to a value of P of less than 0.01. The 25° count, however, was related only to the mean rainfall of the two preceding months ($r = -0.86$, $P = <0.01$). There was no correlation between numbers of bacteria and rainfall for the second or third to last complete months of life.

Tainted samples

Only two outbreaks of bone-taint were reported during this investigation. In the first, four meat samples were taken from each of four sides of tainted beef and a popliteal lymph node from one side. In the second, meat samples and popliteal lymph nodes were obtained from two sides. Aerobic nutrient agar plate counts at 37° were made on the samples taken from the second outbreak. One lymph node had a count of 240/g. and the other node and the meat samples had counts of less than 40/g. In both outbreaks, direct smears from the tainted meat

samples showed numbers of Gram-positive rods. Of 81 isolations obtained from the 21 samples by direct plating, and plating after enrichment in cooked meat medium, 47 were aerobes and 34 anaerobic spore-forming bacteria (clostridia). Seven of the aerobic bacteria were not identified, and eight aerobes and seven anaerobes were discarded as duplicates. The distribution of the remaining 59 different bacteria among the tainted sides is shown in Table I.

Coliform bacteria were found in samples from five of the six tainted sides, *Escherichia coli* being isolated from five samples, *E. freundii* from three and *Aerobacter aerogenes* from one. *Proteus vulgaris* was found in one sample. The aerobic spore-forming bacteria were not as common in the tainted samples as in the ischiatic lymph nodes. *B. cereus* was present in three samples from two sides and *B. pumilus*, *B. circulans*, *B. coagulans*, *B. subtilis* and *B. megaterium* were each found in only one sample. Four types of bacilli were isolated from one sample and two from another sample from the same side. Clostridia were present in 17 of the 21 samples of tainted meat and were differentiated into eight types, the biochemical characteristics of which are shown in Table III. Type 2 was the most common; it was isolated from ten samples and it was present in all the tainted sides. The identity of this *Clostridium* has not yet been confirmed but it appears to differ from the *Clostridia* isolated by Cosnett *et al.*⁸ Type 1 was found in eight samples from three sides and type 3 in four samples from two sides. Only single isolations of the other types were made. Ten samples contained at least two types of clostridia.

When inoculated into fresh meat and incubated at 37° for 20 h., the *E. coli* and *Proteus vulgaris* cultures, and all the bacilli, clostridia and pseudomonad cultures, gave rise to a most unpleasant putrefactive and sour odour similar to that observed in cases of bone-taint. The bacilli appeared to be particularly potent taint-producers.

Table III

Characteristics of clostridium species isolated from tainted beef

Reference No.	1	2	3	4	5	6	7	8
Obligate anaerobe	+	+	—	+	+	+	+	+
Spores	Oval sub-terminal swelling	Oval sub-terminal swelling	Oval terminal swelling	not observed	Oval sub-central swelling	Oval terminal swelling	not observed	not observed
Glucose	—	—	+	+	—	+	—	+
Maltose	—	—	+	+	—	+	—	+
Lactose	—	—	+	+	—	+	—	+
Sucrose	—	—	+	+	—	+	—	+
Mannitol	—	—	+	+	—	+	—	—
Glycerol	—	—	+	+	—	+	—	+
Salicin	—	—	+	+	—	+	—	—
Indole	—	—	—	—	—	—	—	—
Nitrate reduction	—	—	—	—	—	—	—	+
Milk	digestion blackening	digestion blackening	acid clot	acid stormy clot	slow digestion	digestion blackening	acid no clot	acid stormy clot
H ₂ S	—	+	—	—	—	—	+	+
Gelatin								
liquefaction	+	+	—	—	+	—	+	+
Gas produced in cooked meat	+	+	—	—	+	—	+	+

Discussion

The recognition of the importance of quick chilling of the carcass immediately after slaughter has resulted in a much reduced incidence of bone-taint. However, occasional cases do occur, particularly following a deviation from the normal pattern of chilling. This suggests that the taint-producing infection is often present in the carcass and spoilage will result if the conditions are suitable for bacterial multiplication. The present reduction in the number of cases of bone-taint has increased the difficulty of tracing the source and identity of the bacteria concerned.

Some workers regard bone-taint as a special type of spoilage caused by one specific organism or group of organisms. However, the term 'bone-taint' is applied by the meat trade to almost any type of off-odour which has its origin in the deeper tissues, especially those around the hip

joint. The taint may be sweet, sour, putrid or sewer-like. Because of the differences in types of taint, it appears that more than one organism or group of organisms may be involved. This view is supported by the results of this and previous studies in which a number of different organisms have been implicated in taint production. Cosnett *et al.*⁸ isolated 12 Gram-positive rods, anaerobes, sporing and non-sporing aerobes, which they considered to be possible taint-producers. Other workers have implicated several species of bacilli and clostridia as well as other aerobic and anaerobic organisms.¹⁻⁴

In the present study a number of aerobes and anaerobes were isolated from the first four tainted sides. Aerobic bacterial counts made on samples from the second outbreak were low, thus suggesting that anaerobes were responsible for the spoilage in these two sides. As clostridia were isolated from 81% of the samples of tainted meat from both outbreaks, it appears that, in the cases studied, this group of organisms played a major rôle in the development of spoilage. However, as most of the aerobes isolated from the tainted samples also appeared to be capable of causing taint, they may have contributed to the spoilage.

If, according to the theory of Cosnett *et al.*,⁸ the lymph nodes are the centre from which the taint-producing infection spreads into the deeper tissues, it is to be expected that a variety of organisms may be involved. Lymph nodes have been shown to contain a wide range of organisms many of which have been found also in tainted meat. Coliforms, micrococci, achromobacter and pseudomonads were isolated from both tainted meat and lymph nodes. Although bacilli were isolated more often from lymph nodes than from tainted samples, several species were found in both types of sample. Clostridia, including type 2, which was widespread among the tainted samples, were present in a number of lymph nodes.

If conditions in the meat permit bacterial growth, the type of spoilage that develops will depend on the numbers and types of bacteria in the lymph nodes at the time of death, and on factors such as pH, redox potential and temperature of the surrounding tissues during storage. From the number of each type of sample in which clostridia were found during the present investigation, it appears that they were more common in tainted samples than in fresh lymph nodes. Barnes & Ingram¹⁰ have shown that the high redox potential of *pre-rigor* muscle inhibits the growth of *Cl. welchii* by increasing the lag phase. With the onset of *rigor*, the redox potential drops to a level at which this organism can multiply rapidly. Thus, it is to be expected that, in the absence of rapid chilling, samples taken after the onset of *rigor mortis* would contain more clostridia than those collected immediately after slaughter. This emphasises the importance of quick chilling in the reduction of spoilage. To prevent the production of taint by clostridia, the deep-muscle temperature must be reduced sufficiently to restrict bacterial growth, by the time the redox potential has fallen to a level permitting growth. It must be noted that, for animals which are exhausted at the time of slaughter, the *pre-rigor* state and thus the period when growth is inhibited by high redox potential is shorter than normal.¹¹

Although the samples were collected at the same meat works as those of Cosnett *et al.*,⁸ the correlation between rainfall and the percentage of lymph nodes containing Gram-positive rods, observed by these workers, was not found in the present investigation. The proportion of lymph nodes containing Gram-positive rods was about twice that observed by Cosnett *et al.*⁸ This may be due to the use, in the present study, of cultural as well as microscopical methods for the detection of Gram-positive rods. Also, the whole lymph node, rather than just a small portion, was examined.

There was, however, a significant correlation between rainfall and the average number of bacteria in each lymph node, the bacterial load being less after a period of high rainfall than when the rainfall for the two months prior to slaughter had been low. This may be due either to a reduction in the concentration of air- and dust-borne organisms after rain, as suggested by Cosnett *et al.*,⁸ and by Tompkins¹² in respect to the incidence of neonatal tetanus in Nigeria, or it may be related to some nutritional factor leading to variation in resistance to infection. Whatever the actual mechanism by which rain reduces the bacterial load, the observed correlation is evidence that the bacteria found in lymph nodes gained entry into the animal before death.

Although the factors influencing the incidence of potential spoilage organisms in the deeper tissues may not be susceptible to control, it does seem that spoilage can be prevented by chilling the carcass quickly enough to inhibit the multiplication of the spoilage bacteria.

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ISOLATION OF SOME UREA-FORMALDEHYDE COMPOUNDS AND THEIR DECOMPOSITION IN SOIL

By M. I. E. LONG and G. W. WINSOR

The preparation of various members of the methylene-urea series in pure or partially purified form is described, and their analysis and composition is discussed. The rates of mineralisation of the nitrogen of these compounds in soil decreased markedly with increasing molecular size. Methylene-diurea and dimethylene-triurea decomposed too rapidly in the soil to have any practical value as slow-acting fertilisers, whereas a mixture of tetramethylene-pentaurea with higher members of the series was highly resistant to decomposition in soil. Trimethylene-tetraurea proved the most promising compound of the series as a possible constituent of urea-formaldehyde fertilisers, but it appears unlikely that urea-formaldehyde compounds of the methylene-urea type, prepared by condensation under acid conditions, can ever possess the properties of an ideal slow-acting fertiliser.

Introduction

Most of the published work on mineralisation of the nitrogen of urea-formaldehyde compounds in soil is concerned with products prepared by reaction of urea and formaldehyde in acid solution under controlled conditions.¹⁻⁵ The products of reaction have been characterised in various ways, including solubility in water under various conditions,^{1-4, 6} molar ratio of urea to formaldehyde,¹⁻⁴ and content of unreacted urea. In most cases these determinations showed statistically significant relationships with the rates of mineralisation of nitrogen in soil. From the nature of the reactions between urea and formaldehyde, however, it is apparent that almost all the products tested by incubation in soil have been mixtures containing more than one urea-formaldehyde compound, with or without the presence of free urea.

One of the main difficulties in the development of urea-formaldehyde products as nitrogenous fertilisers has been the problem of combining slow, steady mineralisation of nitrogen with a high ultimate 'availability'; thus many of the products tend to decompose rapidly

in the soil at first, whilst materials having lower initial rates of mineralisation were frequently found to contain much of their nitrogen in forms too inert to have any practical value, except perhaps in acid soils.⁷

Studies of pH, urea/formaldehyde ratio and period of reaction in factorial combination showed that careful selection of the reaction conditions gave some improvement in product;⁵ the content of slowly-available nitrogen still remained relatively low, however, and a less empirical approach to the problem seemed desirable, by the isolation and testing of some individual urea-formaldehyde compounds.

The products of reaction between urea and formaldehyde under acid conditions are methylene-ureas (urea units linked by methylene bridges). De Jong & de Jonge⁸ suggested a procedure by which the number of urea units in the molecule can be estimated from the ratio of urea residues to methylene groups:

$$(N_u - 1)/N_u = CH_2/U$$

where N_u = number of urea residues per molecule and CH_2 and U are the molar quantities of methylene groups and urea respectively. As each methylene group links two urea units, the number of methylene bridges will be one less than the number of urea units in the absence of ring formation. This method of calculation has been widely used in the present work; it is particularly effective for the lower members of the methylene-urea series, although increasingly sensitive to experimental error for the larger molecules. Analytical data reported by de Jong and de Jonge lead to values of <7 units of urea per molecule, and indicate that ring-structures do not occur to any appreciable extent.

All the urea-formaldehyde compounds here described have previously been reported in the literature. In several cases, however, where the compound had previously been isolated by fractional precipitation from mixed products a logical synthesis has now been achieved, as indicated in a later section.

Experimental

(a) Analytical methods

The total formaldehyde content of the samples was determined as follows:

The urea-formaldehyde compound (0.2 g.) was distilled with conc. phosphoric acid (25 ml.) and water (40 ml.), 300 ml. of distillate being collected in a receiver containing 2 ml. of 2N-sodium sulphite. Before the end of the distillation the pH of the distillate was adjusted to 8.5 to ensure completion of the reaction between sulphite and formaldehyde. The distillate was then acidified with acetic acid and the determination completed as for free formaldehyde.⁹ It was also shown by analysis⁹ that the methylene-ureas contained no formaldehyde in the form of methylol groups.

Nitrogen was determined by the Kjeldahl procedure.

(b) Preparation of urea-formaldehyde compounds

Methylene-diurea was prepared as described by Kadowaki¹⁰ and purified by recrystallisation from boiling water (m.p. 228–230°) (Found: N, 41.7%; formaldehyde equivalent, 22.6%. $C_3H_8N_4O_2$ requires N, 42.4%; formaldehyde equivalent, 22.7%).

Dimethylene-triurea was prepared by slowly adding 1 mol. of dimethylolurea (12% aqueous solution) to 4 mol. of urea (25% aqueous solution) at 30° and pH 3.0. After 2 h. the precipitate was washed and dried at 40°, to give a product slightly soluble in cold water and melting at ~250° (decomp.) (Found: N, 40.7%; formaldehyde equivalent, 29.0%. $C_5H_{12}N_6O_3$ requires N, 41.2%; formaldehyde equivalent, 29.4%). The preparation of the dimethylolurea for this reaction was based on the method of Baly & Baly,¹¹ urea and formaldehyde at a molar ratio of 1 : 2 plus 5% excess formaldehyde being allowed to interact for 24 h. at room temperature, buffered at pH 8.3. The dimethylolurea was recrystallised from 80% aqueous alcohol and had m.p. 140°.

Trimethylene-tetraurea was prepared as follows: dimethylolurea (2 mol.) and monomethylolurea (1 mol.) were dissolved in 70% aqueous alcohol buffered at pH 4 and maintained at 20°. Precipitation commenced after 30 min., and after 4 h. the precipitate was collected and re-

crystallised from water. The recrystallised product was dissolved in water (2.5% solution) and added to a 25% solution of urea at pH 3 and temperature 40°. The temperature was then lowered slowly to -5° over a period of 10 h. The precipitate was washed with alcohol and air-dried (m.p. ~250° with decomposition) (Found: N, 40.4%; formaldehyde equivalent, 32.8%. $C_7H_{16}N_8O_4$ requires N, 40.6%; formaldehyde equivalent, 32.6%). The monomethylolurea used in the reaction was prepared as already described for dimethylolurea except that the molar ratio of urea to formaldehyde was 1:1 plus 5% excess formaldehyde.

Preparation of *tetramethylene-pentaurea* was attempted as described by Zigeuner *et al.*,¹² based on the method of Dixon¹³ (Found: N, 40.2%; formaldehyde equivalent, 35.3%. $C_9H_{20}N_{10}O_5$ requires N, 40.2%; formaldehyde equivalent, 34.5%). The analytical data indicate that the product was not the pure pentaurea compound, but probably contained a proportion of more highly condensed material (see Discussion). Further separation of the pentaurea compound from higher members of the series was not practicable, and for convenience this material will be referred to in the Tables as 'tetramethylene-pentaurea'.

(c) Mineralisation of nitrogen on incubation with soil

Samples of the various urea-formaldehyde compounds were incubated with 700 g. of moist soil at 23.5° at concentrations equivalent to 300 p.p.m. of N expressed on the basis of oven-dry soil. All treatments were applied in duplicate.

The analytical procedures were as previously described,⁴ except that instead of making separate determinations of ammonia and of nitrate the sum of the two was determined directly by distillation with magnesia and Devarda's alloy.

Results

Mineralisation of the nitrogen of the four methylene-urea compounds was studied in the presence and absence of added calcium carbonate for 26 weeks. The results are given in Tables I and II, together with the pH values at the end of the incubation period.

The data in Table I show that, in the absence of added lime, methylene-diurea decomposed very rapidly in the soil, followed closely by dimethylene-triurea. The nitrogen of trimethylene-tetraurea was mineralised rather less rapidly, but still approached its maximum within 8 weeks at this temperature. In contrast, the 'tetramethylene-pentaurea', representing the higher members of the methylene-urea series, decomposed very slowly in the soil and attained only 15% mineralisation of N within the period of incubation.

Adding calcium carbonate to the soil before incubation markedly retarded mineralisation of N, as previously reported for a range of urea-formaldehyde products.⁷ Methylene-diurea was again found to be quite rapidly mineralised, however. The data for di- and tri-methyleneurea both show a marked lag phase initially, but decomposition was relatively rapid after 1 and 2 weeks, respectively. 'Tetramethylene-pentaurea' again decomposed very slowly in the soil, only 9% being mineralised in 26 weeks as compared with 15% in the unlimed soil.

Discussion

Apart from methylene-diurea, the isolation of individual members of the methylene-urea series in pure form and in quantities sufficient for mineralisation studies proved difficult, and complete separation of the higher members of the series was not achieved, mainly owing to their very low solubility. Becher previously noted¹⁴ the difficulty of isolating the higher methylene-ureas in pure form because of the slight differences in their solubility.

Estimation of the numbers of urea units in the methylene-ureas used in this study, based on the work of de Jong & de Jonge,⁸ gave the following results:

Methylene-diurea	2.02	Trimethylene-tetraurea	4.12
Dimethylene-triurea	2.98	'Tetramethylene-pentaurea'	5.54

The di- and tri-urea compounds appear to have been relatively pure. The calculated value for trimethylene-tetraurea indicates the presence of a small amount of higher members of the series, but in view of the experimental difficulties involved the result is considered acceptable. For 'tetramethylene-pentaurea', however, it is apparent that a considerable quantity of one

or more higher members of the series was present in the product. De Jong & de Jonge⁸ previously concluded that methylene-ureas could contain up to 7 urea units per molecule, with molecular weights in the range 200–500, and the main contaminants are thus likely to be methylene-ureas containing 6 or 7 urea units rather than molecules of appreciably higher chain-length.

Table I

Mineralisation (%) of the nitrogen of some methylene-ureas in a garden soil

Compound	Period of incubation (weeks)								pH after 26 weeks
	1	2	3	4	8	12	16	26	
Methylene-diurea	85	90	90	92	94	92	89	99	5.4
Dimethylene-triurea	71	76	80	82	88	88	88	99	5.4
Trimethylene-tetraurea	44	63	68	73	81	84	81	85	5.9
'Tetramethylene-pentaurea'	2	3	2	3	7	8	10	15	6.7

Table II

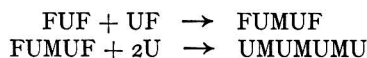
Mineralisation (%) of the nitrogen of some methylene-ureas in a garden soil + 0.5% calcium carbonate

Compound	Period of incubation (weeks)								pH after 26 weeks
	1	2	3	4	8	12	16	26	
Methylene-diurea	58	88	91	90	91	92	85	87	7.6
Dimethylene-triurea	3	60	76	78	81	79	76	82	7.6
Trimethylene-tetraurea	3	8	50	62	69	68	72	67	7.7
'Tetramethylene-pentaurea'	1	2	3	4	5	6	6	9	7.9

As discussed by de Jong & de Jonge,⁸ experimental results for the ratio of methylene bridges to urea residues indicate that ring-structures are not present in methylene-ureas to any appreciable extent. De Jong's method of calculation can make no distinction between branched and straight chains, but information on this point has been obtained by Becher,¹⁴ from infra-red spectra. Dimethylene-triurea and trimethylene-tetraurea were shown to consist of unbranched chains, but the infra-red spectrum of partially purified pentamethylene-hexaurea suggests that branching is probable among the higher methylene-ureas, although there was no evidence for the formation of cyclic structures.

The method used for preparing dimethylene-triurea, though not based on published work, was later found to be similar to that of Kuriyama *et al.*¹⁵ In the preparation of dimethylolurea as an intermediary in the reaction it was considered important to avoid high temperatures (>80°) during recrystallisation; this may explain why the melting point of the dimethylolurea used in the present work (140°), whilst similar to that reported by Baly & Baly (138–140°),¹¹ is higher than those quoted by several other workers including Dixon (~123°),¹³ Scheibler *et al.* (133°)¹⁶ and Zahn & Röchle (126°).¹⁷

As previously noted,⁵ attempts to prepare trimethylene-tetraurea by the method of Kadowaki,¹⁰ involving acidification of methylene-diurea and isolation of the required constituent from the mixed product, were unsuccessful. A stepwise synthesis was therefore devised which may be depicted as:



where F denotes a methylol group, M a methylene group and U a urea molecule or (in a chain) a urea residue or unit.⁸ As previously stated, the product apparently contained a small proportion of higher methylene ureas ($N_u = 4.12$). Becher,¹⁴ following Kadowaki,¹⁰ reported analytical data from which a value of $N_u = 3.97$ may be calculated, in excellent agreement with theory, but the analysis originally reported by Kadowaki gives a lower value ($N_u = 3.68$).

The widest divergences in calculated chain-length (N_u) appear, as might be expected, among the higher methylene ureas. Thus analytical data for a product studied by Becher,¹⁴ referred to as 'pentamethylene-hexaurea' but considered to contain other members of the series, lead to a mean chain length of 5.69 urea residues, little higher than the value of 5.56 obtained in the present work for the product referred to as 'tetramethylene-pentaurea'; both samples are

clearly mixtures. Becher's preparation was based on the method of Kadowaki,¹⁰ and analytical data quoted by the latter worker for the hexaurea compound lead to a value of $N_u = 4.88$, in much closer agreement with tetramethylene pentaurea. Zigeuner,¹² on whose work our 'tetramethylene-pentaurea' sample was based, reported a product whose analysis corresponds to $N_u = 5.14$, in fair agreement with theory.

The data given in Table I show that the rates of mineralisation of nitrogen decrease markedly with increasing chain length, this being particularly so for the impure 'tetramethylene-pentaurea'. Whilst it must be stressed that a proportion of still higher members of the series was undoubtedly present in the latter sample, the mineralisation data show that methylene-ureas of shorter chain length must have been virtually absent and hence that the pentaurea compound itself must be markedly resistant to microbiological decomposition in the soil.

The data in Table II, obtained in a limed soil, show that mineralisation of the di- and tri-methylene-ureas was preceded by periods of relative inactivity, again increasing in duration with chain length. There is little indication of any appreciable proportion of 'slowly available' N under these conditions, however.

The results obtained in this study may explain some of the difficulties in preparing satisfactory slow-acting fertilisers consisting of mixtures of the various methylene-ureas condensed under acid conditions. Thus methylene-diurea is too rapidly decomposed in the soil to have any practical value as a slow-acting fertiliser, and compounds containing more than four urea units in the chain are highly resistant to decomposition in the soil. A certain proportion of N was mineralised relatively slowly from the di- and tri-methylene-ureas, and in the limed soil it would appear that a mixture of these two compounds with methylene-diurea or urea would possess some small reserve of fairly slowly available nitrogen by virtue of the lag phase preceding their decomposition.

On the basis of these results it seems unlikely that methylene-ureas prepared by condensation of urea with formaldehyde under acid conditions could ever possess the properties of an ideal slow-acting fertiliser, at least at the pH values normally encountered in horticultural soils. This does not, of course, imply that existing urea-formaldehyde fertilisers are without value as sources of N for plant growth, but rather that the special advantages sought in this class of fertiliser have not been fully realised. It must also be stressed that the methylene-ureas are only one of the types of compound which can be prepared by reaction of urea and formaldehyde, and that the methylene ethers merit further consideration.

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FLAME-PHOTOMETRIC DETERMINATION OF CALCIUM IN PLANTS

By J. C. BROGAN*

In the flame photometric estimation of calcium, phosphorus and sulphur cause interference which seriously affects the validity of the results. Hitherto it has been necessary to separate these interfering ions, but this is time-consuming. It is now shown that at high concentrations of either phosphate or sulphate ions, the calcium emission is independent of small variations of either anion. This is used as the basis of a flame-photometric method for estimating calcium in plants without separation of interfering ions, but in the presence of excess of sulphate.

Introduction

In connexion with an advisory service to farmers and a field experiment programme on fertility problems, some 15,000–20,000 plant samples are analysed annually for calcium at this laboratory. For analyses on such a large scale, flame photometry seemed a most promising technique, but various interferences had been reported which seriously affect the accuracy of the results.^{1–4} Previously, these interferences were overcome by various separation methods but the methods were all time-consuming,^{3–5} except that of Takahashi & Yoshida,⁶ who reported the use of potassium chloride and sulphuric acid as radiation buffers⁷ in the elimination of phosphate interference.

Consequently, it was decided to examine the interferences normally encountered in the analysis of plants and to attempt to control these without lengthy preparation. The instrument used was a Lange Model 3, incorporating an interference filter at 622 m μ and an air/propane-butane flame.

Experimental

The effects of the alkali metals, sodium and potassium were examined, but these did not interfere significantly and as they had been previously well reported^{3, 8} they were not further considered in this work. Perchlorate ion had been stated to cause positive interference by Baker & Johnson,² but this was not found with our instruments.

Three ions, phosphate, sulphate and aluminium, did cause depressions of the calcium emission and studies on these are recorded below. For each interfering element three series of calcium standards were made up, containing 300, 200 and 100 p.p.m. of calcium as CaCl₂ in water, together with incremental amounts of the ion being investigated. Phosphate was added as KH₂PO₄, sulphate as K₂SO₄ and aluminium as AlCl₃ (aluminium foil dissolved in HCl). A standard solution containing 300 p.p.m. of calcium was used to set the instrument at 100 relative luminosity and all the other standards were read at that setting.

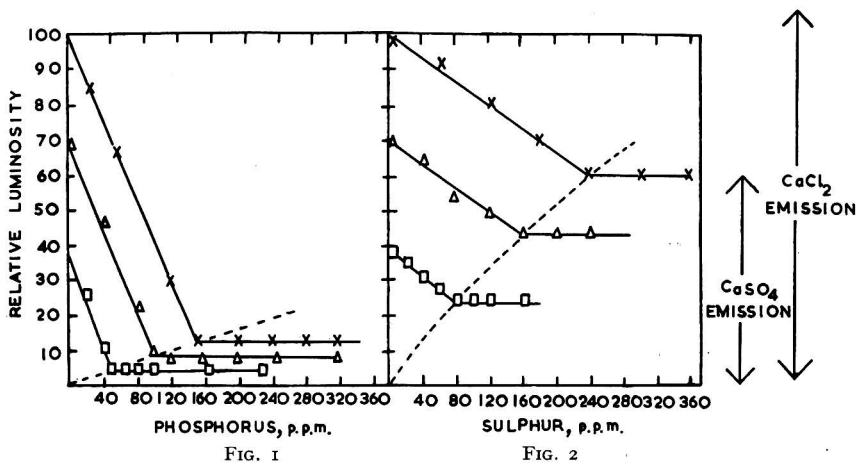
Results

The effect of the concentration of interfering ions on the emission is shown in Figs. 1–3.

From Fig. 1 it can be seen that at high phosphate concentrations the relative luminosity (1) is independent of small variations in phosphate concentration and (2) bears an almost straight-line relationship with calcium concentration shown by broken line (deviations from linearity are a function of photocell used). Similar relationships hold for sulphate and aluminium.

These three ions cause maximum depression of emission at the following ratios of calcium/interfering ion Ca/P 3 : 2, Ca/S 1 : 1 and Ca/Al 1 : 1. A simple explanation of this is the formation in the flame of Ca₃(PO₄)₂, CaSO₄ and a CaAl compound of unknown formula, where only a fraction of the cross-sectional area of the flame is hot enough to cause calcium emission from any of these compounds. It should follow from this, that the relative luminosity at maximum depression would be a function of flame temperature. This was confirmed by comparing two flame types with a series of solutions containing 300 p.p.m. of Ca and different amounts of P as shown in Fig. 4.

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Depression of calcium emission by (Fig. 1) phosphate, (Fig. 2) sulphate
 x Ca 300 p.p.m. Δ Ca 200 p.p.m. □ Ca 100 p.p.m.

In Fig. 4, curve A, the calcium emission with excess phosphate was 40% of that without phosphate, whereas in curve B, with a cooler flame, the calcium emission with excess phosphate was only 12% of the emission in its absence. Similar curves were obtained for sulphate and aluminium. These results were interpreted as evidence that refractory compounds of calcium with the interfering elements were formed in the flame. Presuming that their respective solubility products determine the rates of formation of $\text{Ca}_3(\text{PO}_4)_2$ or CaSO_4 in the flame,³ it would be expected that when both anions were present in the same solution, they would compete for the calcium and the presence of a large excess of one would stop the formation of the calcium salt of the other. The following experiment confirmed this view.

Two series of solutions were made up containing 200 p.p.m. of Ca. The first series also contained 1000 p.p.m. of P as KH_2PO_4 and incremental amounts of sulphate, and the second series 1000 p.p.m. of S as K_2SO_4 and incremental amounts of phosphate. Flame photometric determinations were made as before, the solutions without phosphate or sulphate, respectively, being set at 100 relative luminosity. In all cases readings of relative luminosity of 98-101 were obtained.

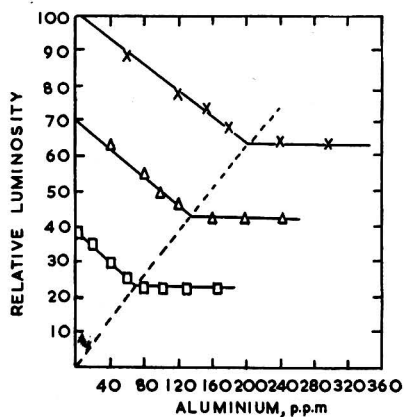


FIG. 3.—Depression of calcium emission by aluminium
 x Ca 300 p.p.m. Δ Ca 200 p.p.m. □ Ca 100 p.p.m.

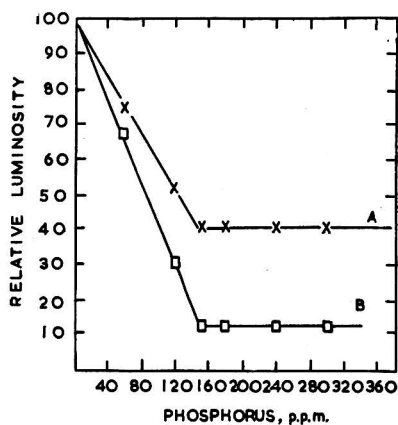


FIG. 4.—Effect of flame on phosphate depression of calcium emission
 A = Oxy/acetylene and Beckmann Model B spectrophotometer at 622 mμ
 B = Air/propane-butane and Lange Model 3 spectrophotometer at 622 mμ

It is concluded therefore that in presence of a large excess of phosphate or sulphate ions, calcium emission is independent of small variations in both.

A similar experiment was carried out to examine interference by aluminium in the presence of a large excess of phosphate or sulphate, in each series the sample with no aluminium being set at 100 relative luminosity. In this case no competition between aluminium and the other ions was observed (Figs. 5 and 6), but the depression of calcium emission by aluminium was reinforced by the presence of phosphate or sulphate. This may point to the formation of complex compounds, e.g. calcium aluminate sulphate in the flame. Interference by aluminium was not further investigated as this element only occurs in most plants (except tobacco) in amounts up to 200 p.p.m. and consequently is not a cause of significant errors.

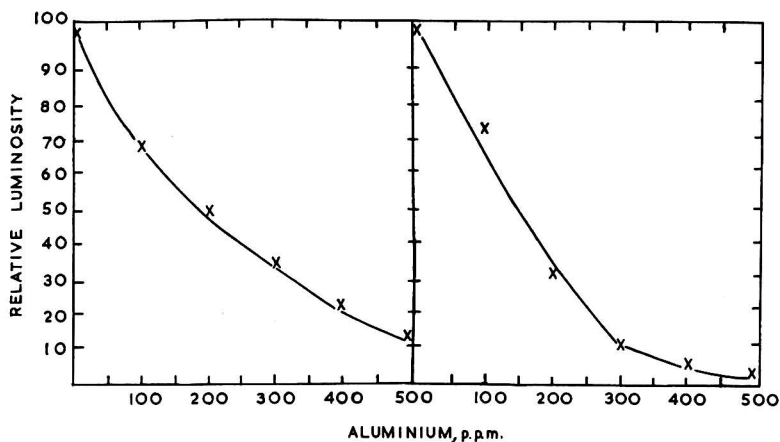


FIG. 5.

FIG. 6.

Interference of aluminium in calcium emission in presence of (Fig. 5) excess phosphate, (Fig. 6) excess sulphate
Ca 200 p.p.m. P or S 1000 p.p.m.

The results reported above suggested a simple flame-photometric procedure for the determination of calcium in plants. This is based on the fact that, after wet oxidation with sulphuric acid, the calcium is present in a solution containing a large excess of sulphate, and its estimation is therefore free from errors due to variations in the P or S content of the plant.

Procedure adopted

Acid digestion: A mixture of conc. HNO_3 , conc. H_2SO_4 and 60% perchloric acid in the ratio 10 : 6 : 3 by volume.

Standard solutions of calcium: Solutions containing 0, 3, 6, 9, 12 and 15 mg. Ca as CaCl_2 in water.

Table I

Calcium content (%) of plant samples as determined by Versenate titration and the flame-photometric method described above

Sample No.	Versenate	Flame photometer	Sample No.	Versenate	Flame photometer
1	1.40	1.40	7	1.48	1.52
2	1.25	1.26	8	2.42	2.36
3	2.36	2.30	9	0.64	0.70
4	1.92	1.85	10	0.68	0.73
5	1.22	1.30	11	0.60	0.63
6	2.08	1.99			

Digest 0.5 g. of dried plant material with 5 ml. of digestion acid over gas burner for 30 min. in a Pyrex Kjeldahl flask graduated at 50 ml. Cool and dilute to volume. Measure the emission at $622 \text{ m}\mu$ in a Lange flame photometer. Treat the standard Ca solutions similarly and prepare a calibration curve.

As the amount of sulphuric acid remaining after digestion varies, a set of calcium standards was made up in strengths of H_2SO_4 varying from 2.5% to 3.5% which is wider than the range normally encountered. No differences were observed at these acid strengths although variations might have been expected due to changes in physical properties of the solutions (viscosity, etc.).

Conclusions

The method was compared with the E.D.T.A. technique of Cheng & Bray⁹ and the results for 10 plant samples are shown in Table I, where the agreement is found to be satisfactory.

The procedure described above has been in use since June 1957 at this laboratory and has given satisfaction over a wide range of plant materials.

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EFFECT OF SOIL TEMPERATURE AND MOISTURE ON THE UPTAKE OF PHOSPHORUS BY OATS

By K. SIMPSON

In a pot experiment with oats as the crop, two levels of soil moisture and two of temperature were used together with five rates of application of superphosphate, on two soils, one high and one low in available phosphorus.

An increase of approximately 5° c in soil temperature maintained throughout the growth period considerably increased the uptake of soil phosphorus from both soils. Lowering of soil moisture tension to field capacity greatly increased the uptake of fertiliser-P from both soils. As found in earlier work, applications of superphosphate to the high-P soil gave rise to the absorption of large 'luxury' amounts of phosphorus with no effect on crop yield.

Introduction

The experiment reported here forms part of a long-term investigation being carried out in the East of Scotland on the effect of phosphate fertilisers on several crops. Earlier work^{1, 2a} showed that there was a positive correlation between the 'apparent' recovery of fertiliser phosphorus by crops and the rainfall during the growing season. It was suggested that the low

'apparent' recovery of fertiliser phosphorus in seasons of low rainfall was the result of an increase in the availability of soil phosphorus brought about by higher soil temperatures, and that the higher soil moisture in wet seasons increased the uptake of fertiliser phosphorus.

The present pot experiment on oats was designed, along with a field experiment on potatoes already reported,^{2b} to examine the above hypotheses. At the same time it was hoped to gain further information about the effects of high applications of superphosphate to crops growing on soils already containing adequate amounts of 'available' phosphorus, in order to supplement previously published work.^{2c, 3}

Experimental

Soils

Two soils were used for the experiment, 'low' and 'high' in available phosphorus (described hereafter as the low-P and high-P soils) with pH values 5.8 and 6.0 respectively. They were sandy clay loams in texture derived from water-worked till of mixed origin. The soils were dug by spade and air-dried in a greenhouse for 3 days before being passed through a $\frac{1}{4}$ -in. screen; 35 lb. of soil were then mixed thoroughly with the same quantity of coarse acid-washed sand and the mixture was sampled for analysis before adding the amounts of fertiliser for a particular treatment. The figures for easily extractable-P were 40 and 140 p.p.m. soluble in 0.2N-HCl, 84 and 207 p.p.m. soluble in 1% citric acid for the low- and high-P soils, respectively. A weighed quantity (14 lb.) of the mixture was then placed in each of five 8-in. fireclay pots previously covered inside with nutrient-free, waterproof bitumastic paint. One central drainage hole in the bottom of each pot was lightly covered with glass wool. This procedure was repeated until 600 pots were filled with soil-sand-fertiliser mixture. Radioactive superphosphate was used and the normal precautions for handling radioactive materials were taken.

Treatments and design

Five rates of application of radioactive superphosphate were used—equivalent to 0, 40, 80, 160 and 320 lb. of P_2O_5 /acre based on 2×10^6 lb. of soil/acre (approximately 0, 3.6, 7.2, 14.4 and 28.8 μ c. per pot). Basal dressings of 56 lb. of N/acre as ammonium sulphate and 56 lb. of K_2O /acre as potassium sulphate were applied to all pots. Sixty pots each of the low-P and high-P soils were subjected to each of the above superphosphate treatments making 600 pots in all. The pots for each treatment were then split into groups of 15 and combined into blocks—giving 75 pots in each of 8 blocks, four of which contained high-P and four low-P soil. Each of the eight blocks consisted of five rows of 15 pots with three replications of each treatment randomised within the rows. The four blocks for each soil were treated throughout the season in the following way:

Block	I	High soil temperature—low moisture tension
	„	II Low soil temperature—low moisture tension
	„	III High soil temperature—high moisture tension
	„	IV Low soil temperature—high moisture tension

Strict control of temperature and moisture was not attempted and the above conditions were achieved by keeping 'high-temperature' pots under greenhouse conditions and 'low-temperature' pots in an open wire cage. Daily soil-temperature records were kept and the difference between the mean daily temperatures for the high- and low-temperature pots was slightly below 5°C. 'Low moisture tension' pots were watered to field capacity daily and the 'high moisture tension' pots were kept relatively dry by watering at intervals of 3 days.

Sowing, sampling and analysis

Oat seeds (Sun II certified stock) were sown at the rate of 70 seeds per pot at a depth of 1 in. below the surface, and all pots were raised to field capacity by placing in shallow pans of water. After this all water was added in the form of aerial sprays.

At the two-leaf stage sufficient plants were removed from each pot by cutting off at the soil surface to leave 50 plants per pot. The oats were sampled at three stages of growth:

(1) the third-leaf stage, (2) the fifth-leaf stage and (3) immediately before 'heading out'. Sampling was carried out by cutting off the plant half an inch above the soil surface. Five pots from each treatment were taken for each sampling, one from each row, and the whole crop was removed. The root portion of the plants was not sampled.

The fresh weight of the 50 plants from each pot was recorded separately and the material dried at 105° for 24 hours after which the dry weight was measured.

Each sample was analysed for total P and ³²P by methods described in a previous paper.^{3a}

Results

In the tables below, the main effects of high and low moisture tension and high and low temperatures are presented. Separate calculations were actually made for all four moisture and temperature conditions, but the effects of soil temperature at high and low moisture tension and the effects of moisture at high and low soil-temperatures were generally similar for most measurements made and the results were therefore pooled to simplify presentation. Although percentage dry matter was measured at all samplings, only the final yield of dry matter is presented as a guide to the effects of treatment and uptake on yield.

The effect of treatments on the total uptake of P₂O₅ in mg. per 50 plants is given in Table I.

Table I

Effect of treatments on total uptake of P₂O₅ by oats at three stages of growth

Treatment	P ₂ O ₅ applied lb./acre	Total uptake of P ₂ O ₅ , mg./50 plants					
		High-P soil			Low-P soil		
		1st sampling	2nd sampling	3rd sampling	1st sampling	2nd sampling	3rd sampling
Low moisture tension	0	42.0	77.1	117.2	11.3	10.5	19.5
	40	42.5	96.6	137.6	9.5	14.5	20.1
	80	56.9	115.4	168.5	13.3	21.8	22.4
	160	55.9	123.4	193.1	15.1	43.5	50.1
	320	92.6	161.7	232.9	32.8	63.1	68.9
High moisture tension	0	30.0	57.9	126.1	7.9	14.9	23.4
	40	39.7	81.7	126.9	10.7	16.5	28.7
	80	43.0	82.1	131.5	11.8	17.8	31.3
	160	56.9	87.0	144.5	16.3	32.2	42.9
	320	72.3	131.5	148.8	23.9	56.0	85.9
High soil temperature	0	43.8	80.5	128.4	13.0	16.3	26.5
	40	43.2	103.0	141.2	10.9	18.9	27.8
	80	54.3	114.7	155.0	15.1	22.7	29.9
	160	56.8	113.8	172.8	18.9	38.5	47.6
	320	79.2	158.1	210.4	28.8	54.3	70.6
Low soil temperature	0	28.2	55.5	114.9	6.2	9.1	16.4
	40	39.0	75.3	123.3	9.3	12.1	21.0
	80	45.6	82.5	145.0	10.0	16.9	23.8
	160	56.0	96.6	165.1	13.8	37.3	45.4
	320	85.7	131.5	171.3	28.0	64.8	84.2
Means of all treatments							
Low moisture tension		58.0	114.8	169.9	16.4	30.7	36.2
High moisture tension		48.4	88.0	135.6	14.1	27.5	42.4
High temperature		55.5	114.0	161.6	17.3	30.1	40.5
Low temperature		50.9	88.3	143.9	13.5	28.0	38.2
L.S.D. (P = 0.01)		4.5	11.1	15.3	1.9	3.0	4.0

The total uptake of P from the high-P soil was considerably increased at low moisture tension in all fertiliser-treated pots, particularly at the later samplings. At the final sampling the increase was 9, 28, 34 and 57%, respectively, of the uptake at high moisture tension, for the 40, 80, 160 and 320 lb. of P₂O₅/acre rates of application. This effect was not found on the low-P soil where, at the lower rates of application (40 and 80 lb. of P₂O₅/acre), a reduction of moisture tension tended to decrease total P uptake by the plants at the final sampling.

Increases in soil temperature on both soils, generally gave rise to increased uptake of P except at the highest level of application throughout the growth period on the low-P soil and on the high-P soil at the first sampling.

Although the total uptake of P increased with rising levels of fertiliser-P application, the curves which may be drawn from the data in Table I, particularly for the low-P soil at high moisture tension and low soil temperature, followed the lower part of a sigmoid curve, showing no tendency to reach a maximum even at the highest level of application of superphosphate. This effect was shown to a very reduced extent on the low-P soil at low moisture tension and high soil temperature and was absent on the high-P soil.

Table II shows the effect of treatments on the uptake of fertiliser phosphorus (^{32}P).

Table II

Effect of treatments on uptake of fertiliser P_2O_5 by oats at three stages of growth

Treatment	P_2O_5 applied, lb./acre	Uptake of fertiliser P_2O_5 , mg./50 plants					
		High-P soil			Low-P soil		
		1st sampling	2nd sampling	3rd sampling	1st sampling	2nd sampling	3rd sampling
Low moisture tension	0	—	—	—	—	—	—
	40	5.5	10.1	13.3	1.1	3.3	4.3
	80	14.3	21.2	37.1	4.0	7.6	7.7
	160	20.0	40.6	58.4	6.9	22.7	27.1
	320	53.2	78.7	121.9	21.3	55.1	51.9
High moisture tension	0	—	—	—	—	—	—
	40	5.4	9.8	11.7	1.1	2.6	4.5
	80	8.9	17.8	24.1	2.1	4.1	8.8
	160	21.9	32.3	40.0	5.4	16.1	18.7
	320	39.2	63.0	63.6	14.1	34.0	43.4
High soil temperature	0	—	—	—	—	—	—
	40	5.8	10.8	13.5	1.1	2.9	4.5
	80	12.9	23.4	27.1	3.8	5.8	7.9
	160	21.3	39.4	52.6	6.5	18.7	21.4
	320	44.3	79.1	98.2	17.0	35.2	40.2
Low soil temperature	0	—	—	—	—	—	—
	40	5.1	9.1	11.5	1.1	2.9	4.2
	80	10.4	15.8	24.1	2.3	5.9	8.6
	160	20.7	33.5	45.7	5.7	20.1	24.4
	320	48.1	62.6	87.3	18.5	49.9	55.0
Means of all treatments							
Low moisture tension		23.3	37.7	57.7	8.3	22.2	22.8
High moisture tension		18.9	30.7	34.9	5.7	14.2	18.9
High temperature		21.1	38.2	47.9	7.1	15.7	18.5
Low temperature		21.1	30.3	42.2	6.9	19.7	23.1
L.S.D. ($P = 0.01$)		3.2	4.6	5.0	1.1	1.9	2.3

The uptake of fertiliser-P increased linearly with the rate of superphosphate application in the high-P soil whatever the moisture or temperature conditions. On the low-P soil, however, the increase in uptake with fertiliser application followed the lower part of a sigmoid curve as in the total uptake, and was not tending to a maximum at the 320 lb. of P_2O_5 /acre rate of application.

Fertiliser-P uptake was increased at low soil moisture tension, particularly at higher rates of application, at all sampling times. This effect was most marked in the low-P soil at the second sampling after which stage the plants at low moisture tension ceased, whilst the plants grown at high moisture tension continued to absorb fertiliser-P. In the high-P soil, however, the increases in fertiliser-P uptake under low moisture tension were striking at all three samplings, and the difference between uptake under low and high moisture tensions increased as the season progressed. At the third sampling the uptake under low moisture tension was 14, 54, 46 and 91% greater than that under high moisture tension for the 40, 80, 160 and 320 lb. of P_2O_5 /acre treatments respectively.

On the high-P soil, the uptake of fertiliser-P was increased by increasing the soil temperature. The effect was not so marked as that of soil moisture and was not found at the first sampling. Soil temperature had little effect on the uptake of fertiliser-P from low-P soil except at the higher levels of superphosphate application (160 and 320 lb. of P_2O_5 /acre) where an increase in soil temperature decreased the uptake.

The uptake of fertiliser-P, throughout the growth period, was very much lower on the low-P soil than on the high-P soil. At the first sampling (low moisture tension) it varied from one-fifth at the 40-lb./acre rate to two-fifths at the 320-lb. rate. At the third sampling the margin was narrower—one-third at the 40-lb. rate and almost half at the 320-lb. rate.

Table III shows the effect of treatments on the uptake of soil-P.

Table III

Effect of treatments on uptake of soil-P by oats at three stages of growth

Treatment	P_2O_5 applied, lb./acre	Uptake of soil P_2O_5 , mg./50 plants					
		High-P soil			Low-P soil		
		1st sampling	2nd sampling	3rd sampling	1st sampling	2nd sampling	3rd sampling
Low moisture tension	0	42.0	77.1	117.2	11.3	10.5	19.5
	40	37.0	85.5	124.4	8.4	11.3	15.8
	80	42.6	94.1	131.4	9.3	13.2	14.7
	160	35.8	82.8	134.7	9.8	20.9	23.0
	320	39.4	83.0	111.0	11.5	22.0	17.2
High moisture tension	0	30.0	58.9	126.1	7.9	14.9	23.4
	40	34.4	71.9	115.2	9.6	13.9	24.2
	80	34.1	64.3	107.5	9.7	13.7	22.5
	160	35.1	60.7	104.7	10.9	16.1	24.2
	320	33.2	64.8	85.3	9.8	22.0	42.5
High soil temperature	0	43.8	80.5	128.4	13.0	16.3	26.5
	40	37.4	92.1	127.7	9.8	16.0	23.3
	80	41.4	91.6	128.0	11.3	16.9	22.0
	160	35.5	74.3	120.0	12.4	19.8	26.2
	320	35.0	79.1	112.2	11.8	19.1	30.4
Low soil temperature	0	28.2	55.5	114.9	6.2	9.1	16.4
	40	34.0	66.3	111.9	8.2	9.2	16.7
	80	35.3	66.8	110.9	7.7	10.0	15.2
	160	35.4	63.2	109.4	8.3	17.2	21.0
	320	37.6	68.7	84.1	9.5	24.9	29.3
Mean of all treatments							
Low moisture tension		39.4	84.5	123.7	10.1	15.6	18.0
High moisture tension		33.4	64.1	107.8	9.6	16.1	27.4
High temperature		38.6	83.5	123.3	11.7	17.6	25.7
Low temperature		34.1	64.1	106.2	8.0	14.1	19.7
L.S.D. ($P = 0.01$)		4.5	8.7	12.3	1.3	2.0	2.9

The soil-P uptake from untreated pots on the high-P soil was, as expected, between 3.4 and 4.5 times as great as that from the low-P soil.

The figures shown in Table III leave no doubt about the effect of soil temperature in increasing the uptake of soil-P by the plant. The effect was most marked on the low-P soil where the uptake of soil-P was increased from 6.2 to 13.0 mg. per 50 plants at the first, from 9.1 to 16.3 at the second, and from 16.4 to 26.5 at the third sampling—increases of 109, 79 and 62%, respectively. The corresponding increases for the high-P soil were 55, 45 and 12%.

On the high-P soil and at low rates of application (40 and 80 lb.) on the low-P soil, particularly at the higher soil temperature, the addition of superphosphate tended slightly to depress the uptake of soil-P.

Lowering the soil moisture tension increased the uptake of soil-P from control pots of both soils at the first and second samplings but lowered it slightly at the final sampling, the reduction being 20% and 8% on the low- and high-P soils, respectively.

There was little stimulation of uptake of soil-P by addition of superphosphate except at the highest level of application (320 lb./acre) on the low-P soil. In fact, at high moisture tension (final sampling) on the high-P soil, each increase in superphosphate application further depressed the uptake of soil-P.

Discussion

Effects of superphosphate application

The uptake of total and fertiliser phosphorus was increased steadily under all temperature and moisture regimes by increasing application of superphosphate. On the high-P soil the increases were linear for fertiliser phosphorus. This agrees with previous findings by Verma *et al.*^{3a} The low-P soil behaved differently, only very small increases in fertiliser-P uptake being brought about by applications of 40 and 80 lb. of P_2O_5 /acre, and greater increases by the higher levels (160 and 320 lb.). This effect was accompanied by reductions in soil-P uptake at the lower levels of application and a corresponding reduction in final crop yield at these levels where the soil was kept at low moisture tension or high temperature. The sigmoid nature of the curves for total and fertiliser-P uptake and yield obviously becoming more marked in a wet or warm soil indicated the strong fixing power of this soil which has been overcome only by large applications of soluble P. The decreases in soil-P uptake, not accompanied by considerable increases in fertiliser-P uptake, on the low-P soil are not easy to explain.

Yield figures at the various sampling times have not been presented in full as the main object of this study was to investigate P uptake as affected by soil moisture and temperature, but a few salient features are of interest.

Superphosphate treatments had no significant effects on yield in the high-P soil at any stage of growth. On this soil the mean yield of dry matter at the final sampling was 29.3 g. per 50 plants. There was, in fact, some slight indication of depressions in yield under high temperature and low moisture tension at the highest level of application, associated with the uptake of very considerable amounts of fertiliser-P. For example an extra uptake of 122 mg. of P_2O_5 per 50 plants at the final sampling—doubling the control uptake of 120 mg.—gave no increase in yield, confirming previous work.^{3a,b} This luxury uptake has been found to produce toxic effects^{2b} and further work will be carried out as to the form in which this excess P occurs in the plant tissue. The relative P-supplying power of the two soils as indicated by 'readily soluble' estimations, P uptake from control plots and yield is shown in Table IV.

Table IV

Comparison of yield, phosphorus uptake and soil analysis data

	High-P soil	Low-P soil	Ratio	$\frac{\text{high P}}{\text{low P}}$
0.2N-HCl-soluble P_2O_5 , p.p.m.	140	40		3.5
1% citric-acid-soluble P_2O_5 , p.p.m.	207	84		2.5
Total P_2O_5 uptake from control, mg./50 plants	122	22		5.5
Yield (control), g. of dry matter/50 plants	30.0	6.8		4.2

Thus the control yield ratio is in fair agreement with the soil analysis data but there is some indication that the high-P soil has supplied relatively more P to control plants than was indicated by the soil analysis results.

Effects of moisture regimes

The final total P uptake from control pots (Table I) was decreased at low moisture tension. This effect was more marked on the low-P soil where the decrease amounted to 17% as compared with 8% on the high-P soil. It has been previously noted that control uptake of phosphorus in wet seasons was lower than that in dry ones.^{1, 2a}

Tables I and II leave little doubt about the increase in total and fertiliser-P uptake which occurred in the plants grown on soils kept at field capacity. This effect was most marked in the early season and was maintained throughout the season on the high-P soil, but not at the final sampling on the low-P soil. The inference is that high moisture tension, particularly

on the low-P soil, while decreasing the rate of absorption of P in the early part of the season also slowed down the rate of fixation—the rate of absorption at high moisture tension between the second and third samplings is much higher at the 40- and 80-lb. levels where fixation of fertiliser-P is most likely to have a marked effect.

A high degree of fixation of fertiliser-P on the low-P soil is suggested by the comparative uptake of fertiliser-P from the two soils. At the first sampling approximately one-fifth as much fertiliser-P had been absorbed from the 40-lb./acre treatment and two-fifths from the 320-lb. rate on the low-P soil compared with the high-P soil.

The actual effect of moisture tension on phosphorus absorption is best demonstrated by Table V with data extracted from Table I.

Table V

Net extra uptake of total phosphorus (mg./50 plants) at two levels moisture tension
(High-P soil)

P ₂ O ₅ applied, lb./acre	0	40	80	160	320
Low moisture tension	—	20.4	51.3	75.9	115.7
High moisture tension	—	0.8	5.4	18.4	22.7

It is obvious from Tables I, II and V, that the chief effect of lowering soil moisture tension was a striking increase in the uptake of fertiliser-P. This effect was present throughout the season with both soils and was reflected in a considerable yield increase in the high-P soil from 23.8 to 34.8 g. dry matter per 50 plants as a mean of all treatments. Jordan *et al.*⁴ and Simpson,^{2c} working with potatoes, have previously found that the lowering of soil moisture tension increased the uptake of fertiliser-P. On the low-P soil the extra fertiliser-P absorbed resulted in little yield increase as it was accompanied by considerable decreases in uptake of soil-P. The uptake of soil-P (Table III) from the high-P soil was considerably depressed by increasing applications of fertiliser-P in the later part of the season. This agrees with previous findings with both potatoes and oats,^{2a, 3a-c} and with those of other workers,⁵ but disagrees with the findings of Strzemienski⁶ and Woltz *et al.*,⁷ who quoted considerable stimulation of soil-P uptake by applications of superphosphate. The soil-P uptake was also depressed by the lower rates of application (40 and 80 lb./acre) on the low-P soil but was slightly increased by higher rates. This effect is not easy to explain. Whatever the cause, the effect was directly reflected in the yield curves.

Effects of soil temperature

While, as seen above, the effect of varying soil moisture was observed mainly in the uptake of fertiliser-P, there is no doubt that changes in soil temperature affected the uptake of soil-P. Reference to Table III shows that the 5° increase in soil temperature more than doubled the uptake of soil phosphorus on the low-P soil control pots at the first sampling, with corresponding increases of 79 and 60% at the 2nd and 3rd samplings. A similar effect though much smaller was seen in the high-P soil. This effect was reflected in higher control yields. This is in full agreement with the higher control yield found in previous field experiments on phosphate-deficient soils in warm seasons.^{2a}

It seems reasonable to suggest that mineralisation of organic P has played a part in the increased availability of soil phosphorus. Several workers, including Eid,⁸ Hayashi & Takijima,⁹ and Jackman & Black,¹⁰ have established that mineralisation of organic-P takes place readily in the soil and that the process is considerably speeded up by increases in soil temperature.

Increased soil temperature also raised the uptake of fertiliser and consequently of total P on the high-P soil but it had little effect on the low-P soil.

Table VI shows that the extra uptake of P brought about by superphosphate applications (compared with the uptake from controls) is higher at the lower soil temperature. These were the data formerly used to calculate 'apparent recovery' of fertiliser phosphorus which would, therefore, be lower at the high soil temperature.

This is in agreement with the author's previous suggestion that low 'apparent recoveries' of fertiliser-P by crops in warm seasons are related to increased availability of soil phosphorus.^{2a}

This effect was not observed on the high-P soil and it must be concluded that, in this soil the main effect was that of a stimulation of fertiliser-P uptake by reducing moisture tension,

Table VI

Effect of soil temperature on the net extra uptake (mg./50 plants) of phosphorus at different levels of application

(low-P soil, 3rd sampling)				
P ₂ O ₅ applied lb./acre	40	80	160	320
High temperature	1.3	3.4	21.2	44.1
Low temperature	4.6	7.4	29.0	67.8

with temperature effects playing a less important part. In the low-P soil, however, while increasing soil temperature had little effect on the uptake of fertiliser-P (if anything a depression), it had a remarkably stimulating effect on soil-P uptake.

Conclusions

(1) An increase of approximately 5° in soil temperature gave rise to considerable increases in the uptake of soil-P by oats, particularly on a soil low in available P.

(2) Increased temperature also raised the uptake of fertiliser-P from the high-P soil and of total P from both soils.

(3) Lowering soil moisture tension by keeping soils at field capacity greatly increased the uptake of fertiliser-P from both soils, but there were indications that the rate of fixation of fertiliser-P was decreased at high moisture tension as the crops grown under these conditions continued to absorb P at a higher rate in the later part of the season.

(4) Soil-P uptake was depressed increasingly by each increment of superphosphate application on the high-P soil and by the 40- and 80-lb. rates of P₂O₅/acre on the low-P soil.

(5) Yield was unaffected by superphosphate treatments on the high-P soil despite high levels of uptake of fertiliser-P and was depressed by 40 and 80 lb. of P₂O₅/acre and increased by 160 and 320 lb. of P₂O₅/acre on the low-P soil.

(6) The results confirm previous suggestions that temperature and moisture effects were responsible for low 'apparent recovery' of fertiliser-P in warm dry seasons as compared with cool wet ones.

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KAFFIRCORN MALTING AND BREWING STUDIES.**V.*—Occurrence of β -Amylase in Kaffircorn Malts†**

By L. NOVELLIE

Contrary to commonly accepted views, sorghum (kaffircorn) malts have been found to contain β -amylase in considerably more than traces, 18–39% of the saccharifying activity being due to the β -amylase. The α - and β -amylases have been found to develop at approximately the same rate during germination. Sorghum β -amylase has been prepared free from the α -enzyme and its properties studied for the first time. Since the β -amylase of sorghum closely resembles those of barley malt and wheat, the low diastatic power of kaffircorn malts compared with those of wheat and barley is attributed only to the lower proportion of β -amylase and not to any fundamental difference in its properties.

Introduction

Barley, wheat and rye are rich in β -amylase in the ungerminated state, malting serving argely to solubilise and activate the enzyme. Maize, rice and sorghum, on the other hand, are practically devoid of amylase activity in the ungerminated state,^{1, 2} germination supposedly resulting in the formation of α -amylase only.

The literature on the malting of sorghum and the properties of the sorghum amylases is particularly scanty in comparison with that on barley, wheat and rye. Sorghum malts are commonly regarded as containing large quantities of α -amylase with such minute amounts of β -amylase that detection is difficult even with sensitive methods of estimation.^{1, 3} Various South African kaffircorn malts of commerce analysed in this laboratory were found, however, to contain a considerable proportion of β -amylase. Since kaffircorn malts have a low diastatic activity and since a high proportion of adjunct to malt is used in kaffir beer brewing, diastatic power is a critical factor. The discovery of the presence of β -amylase in the malt was, therefore, of particular interest and importance and a study has been made of the formation of the enzyme during malting and of its properties when purified.

Studies on the purification of the cereal amylases are relatively few and have been mainly concerned with the separation of the α - and β -enzymes from one another. Once this has been accomplished, little further purification has been attempted, especially with modern enzyme techniques.^{4, 5, 6} Noteworthy exceptions to this are provided by the work of Meyer and co-workers^{6–8} and Balls and co-worker.⁹ The former have crystallised wheat and malt β -amylases and the latter malt α -amylase. The wheat β -amylase was found to be electrophoretically homogeneous but was not examined in the ultracentrifuge. The homogeneity of the β -amylase from barley malt was not determined. An important contribution to the question of the homogeneity of this enzyme has been made by Pollock and co-workers^{10, 11} who obtained a number of fractions with similar β -amylase activity but with different electrophoretic mobilities. These electrophoretic fractions showed inhomogeneity in the ultracentrifuge. None of the cereal β -amylases has therefore been obtained in a homogeneous state as defined by both electrophoretic and ultracentrifuge analysis.

Wheat, barley and malt are well known to be richer in β -amylase than the other cereals or their malts and are thus ideal sources of the enzyme. The purification and isolation of a β -amylase from a cereal or malt poor in this enzyme is more difficult and does not appear to have been previously attempted.

Experimental*Malt*

Typical commercial malts from a number of maltsters were used in the determination of the proportion of β - to α -amylase to be found in kaffircorn malts and in the purification of the β -amylase.

* Part IV: *J. Sci. Fd Agric.*, 1960, **11**, 408

† Part of this paper was read at the 13th Annual Convention of the South African Chemical Institute, July 1959

To follow the development of β -amylase during germination, malts were prepared in the laboratory from Barnard's red kaffircorn. The grain was germinated² at 30° for 6 days, samples being taken from the second day of germination onwards.

Determination of the ratio of β - to α -amylase

The method used was that of Preece.¹² The method of determining diastatic power was, however, replaced by that specially developed for kaffircorn malts.¹³ Determination is made of the joint α - and β -amylase activity (diastatic power) and then of the α -amylase activity after the β -amylase has been destroyed by heating to 70° in the presence of calcium ions. After correction for the small concomitant loss of α -amylase, the β -amylase activity is found by difference.

Purification of sorghum β -amylase

Malt was extracted (overnight) with water (malt : water, 6 : 10 w/w) containing toluene (5% v/v). The total amylase activity of the filtered extract was precipitated by addition of ammonium sulphate to 65% of saturation and the precipitate, plus 3–5 volumes of water, dialysed in the cold until free of salt. The dialysed solution was cooled to 0–2° and brought to pH 3.2–3.4 by the addition of acetic acid (5N), and after half an hour, adjusted to pH 5 by the addition of solid sodium acetate or of sodium hydroxide (0.5N). Denatured material was centrifuged, and the supernatant crude β -amylase solution was dialysed and fractionated with ammonium sulphate, the bulk of the activity precipitating at 45–65% of saturation. After dialysis, this material was fractionated with alcohol at –10° to –12°, the precipitate obtained at an alcohol concentration between 33 $\frac{1}{3}$ and 66 $\frac{2}{3}$ % v/v being spun down in a refrigerated centrifuge after 20 min. After being taken up in ice water, the precipitate was dialysed in the cold. Further purification of the β -amylase was achieved by adsorption on calcium phosphate gel prepared according to Keilin & Hartree.¹⁴ As the quantity of gel required for the fractionation varied from one preparation to another, the amount was determined by a pilot experiment in each case. The adsorbed β -amylase was eluted as soon as possible with ammonium sulphate solution (1/10 saturated), and the eluate dialysed against sodium chloride (0.5% w/v) or, for electrophoresis or ultracentrifugation against a suitable buffer, until free from ammonium ions.

Examination of purified fractions

The small amounts of α -amylase left after acid treatment were estimated by the dextrinisation time method,¹³ using extended reaction periods.

Total amylase activity (or β -amylase activity when α -amylase was absent).—This was determined by the diastatic power method specially developed for kaffircorn malts.¹³ Specific activity was calculated as K.D.U./mg. of protein N (see 13).

Protein nitrogen.—After dialysis to remove non-protein nitrogen, nitrogen was determined in the solutions by a modification of the method of Ma & Zuazaga¹⁵ with a mercury catalyst and a 4-h. digestion period.

Ultracentrifugal analysis.—Sedimentation diagrams were obtained on solutions in 0.1M-acetate buffer, pH 4.7, in a Spinco model E ultracentrifuge at 56,000 r.p.m.

Results and discussion

Ratio of β - to α -amylase in sorghum malts.

A selection of results obtained with commercial malts is given in Table I. This shows that β -amylase is present in much more than traces, 18–39% of the saccharifying activity of the malts being due to the β -enzyme. The ratio of β - to α -amylase varied from 0.22 : 1 to 0.64 : 1. Although the ratio found in birdproof kaffircorn malt is somewhat higher than those found for the normal types of malt, it is clear that there is no fundamental difference between the two types as far as their amylase composition is concerned. The ratio of β - to α -amylase remained practically constant throughout malting (Table II) showing that the two amylases developed at the same rate.

Preece¹⁶ found that the ratio of β - to α -amylase in barley malts varied from 2.5 : 1 to 6 : 1

(average about 4:1), and was approximately 1:1 for oat malts.^{16b} Bose & Krishna¹⁷ reported the ratio to be 0.25:1 for ragi (*Eleusine coracana*) malts. Sorghum malts are therefore very poor in β -amylase compared with barley malts, and show a closer relationship to oat and ragi malts.

Table I

Ratio of β - to α -amylase in commercial malts		
Malt	β -Amylase as % of total amylase	β -/ α -amylase ratio
Normal type (soluble amylases)		
1	34	0.52:1
2	18	0.22:1
3	28	0.39:1
4	29	0.41:1
5	23	0.30:1
Birdproof kaffircorn malt		
M55/11	39	0.64:1

Table II

Influence of length of germination on the ratio of β - to α -amylase in short red variety of kaffircorn			
Days of germination	Diastatic power, K.D.U./g.	β -Amylase % of total amylase	Ratio of β -/ α -amylase
2	23.0	26	0.35:1
3	38.5	30	0.43:1
4	54.2	27	0.37:1
5	73.5	28	0.39:1
6	72.8	26	0.35:1

Purification of sorghum β -amylase

The low proportion of β -amylase in sorghum malts made it necessary to prepare large volumes of malt extract of as high a concentration as possible. Because of the difficulty in handling such volumes, it was found essential first to precipitate the amylases with ammonium sulphate to effect concentration of the amylases and after removal of all but traces of α -amylase by treating the dialysed ammonium sulphate precipitate with acid, it was possible to start the actual purification procedures. A variety of fractionation procedures was tried; those finally chosen are outlined in Table III. It was not found possible to eliminate the α -amylase completely by acidification: in all cases traces remained except at higher temperatures when the loss of β -amylase became too great to make the method practicable. The remaining α -amylase was, however, removed by the succeeding steps of purification. The preparation obtained from the alcohol precipitation produced 42 mg. of maltose/min./mg. of N from starch but was incapable of dextrinising the starch to a red-brown iodine colour even after action for 72 h. The β -amylase had therefore been completely freed from α -amylase at this stage.

Table III

Purification of sorghum β -amylase			
Stage	Total activity K.D.U.	Specific activity K.D.U./mg. N	
		(α + β)-amylase	β -amylase
0 (Malt extract)	~260,000	4-5	~1*
1 Ammonium sulphate precipitation to 65% saturation and dialysis	258,108	46	~9*
2 Acidification to remove α -amylase	49,852	—	7.7
3 Ammonium sulphate fractionation at pH 5, precipitate at 45-65% saturation dialysed	29,112		54.1
4 Alcohol fractionation 33 $\frac{1}{2}$ -66 $\frac{3}{8}$ v/v	24,916		90
5 Calcium phosphate adsorption eluate from:			
(a) Pilot expt.			984
(b) Bulk, 3rd adsorption step	4254		496
**4th " "	5120		453
	9374		

* Calculated on a β -/ α -amylase ratio of 1:4

** This fraction was used for the ultracentrifuge analysis

The results in Table III show that the β -amylase was purified approximately 500-fold in the bulk experiments, and the yield was 3-4% of the original diastatic activity. A considerably more active fraction than the bulk material was obtained in one pilot experiment. This had

an activity of 984 K.D.U./mg., i.e., 1000-fold that of the starting material. To compare the activities of these preparations of sorghum β -amylase with those of wheat and malt β -amylases made by Meyer *et al.*,⁷ they were calculated in terms of Meyer's units using a factor of 0.5 for converting activities at 30° to 20°. These gave values of 1035 and 2550 units per mg. of N compared with 1450 and 1660 units for wheat and malt β -amylases, respectively. The activity of the sorghum enzyme is therefore in no way inferior to those of the other cereal β -amylases.

Electrophoresis of sorghum β -amylase on paper showed only one band. In the ultracentrifuge, however, two peaks were visible, one of extreme sharpness (Fig. 1); the corrected sedimentation constants for these were 2.2 and 3.3 Svedberg units. It is interesting to note that Cooper & Pollock¹⁰ found sedimentation constants of 2.1, 2.3 and 3.4 for some of their barley malt β -amylase fractions. Although the activity of the sorghum β -amylase was comparable with that of other cereal β -amylases, it is clear that further work is needed to obtain a completely homogeneous preparation of the enzyme.

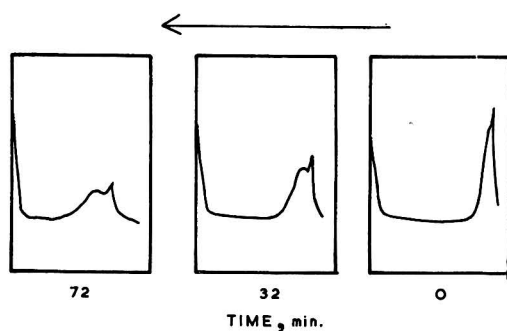


FIG. 1.—Sedimentation diagram for sorghum β -amylase

Properties of purified sorghum β -amylase

Variation of activity with pH.—The variation of the activity of sorghum β -amylase with pH at 30° is shown in Fig. 2. Optimum activity was found at pH 5.3–5.4, although, the peak being broad, activities within 5% of the maximum are encountered in the pH range 4.6–6.6. Meyer and co-workers found the optimum pH for β -amylase activity of barley malt was 4.8–5.7 and of the wheat enzyme 4.8–6.5, with maxima at pH 5.2 and 5.3 respectively.^{7, 8} The form of the pH/activity curve for sorghum β -amylase more closely resembles that for wheat β -amylase than that for the barley malt enzyme.⁷ There is nevertheless considerable similarity between the three enzymes with regard to the effect of pH on their activity.

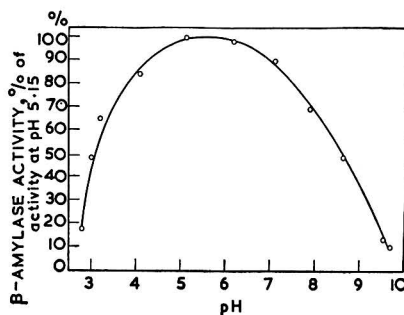


FIG. 2.—Sorghum β -amylase activity as a function of pH at 30°

Amylase stability and pH.—The effect of pH on the stability of the β -amylase was determined at 30° by keeping the enzyme at various pH values for 4 h. Optimum stability was at pH 5.5 (Fig. 3). Wheat and malt β -amylases showed considerable stability between pH 4 and 8 at a temperature of 20°. The rather poor stability of sorghum β -amylase was probably due to the use of a higher temperature, 30°, and of very dilute enzyme solutions in the tests.

Activity and temperature: energy of activation.—The activity of the amylase was determined at 10°, 20°, 30° and 40°. The temperature coefficients calculated from the results were 2.26 for the range 10–20°, 1.97 for 20–30° and 1.80 for 30–40°. The energy of activation, obtained from the plot of log (velocity) versus reciprocal of the absolute temperature, was 12,070 cal./mole between 10° and 40°. This is close to that for wheat β -amylase, viz., 13,000 cal./mole between 10° and 20°, and 9300 cal./mole between 20° and 40°. The Arrhenius plot (Fig. 4) for sorghum β -amylase is, however, a straight line between 10° and 40°, unlike the plots for wheat and malt β -amylases which show a break in the vicinity of 20°.

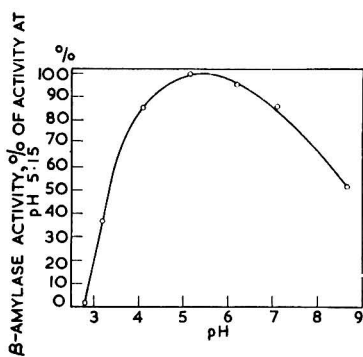


FIG. 3.—Stability of sorghum β -amylase at various pH values (held at 30° for 4 h.)

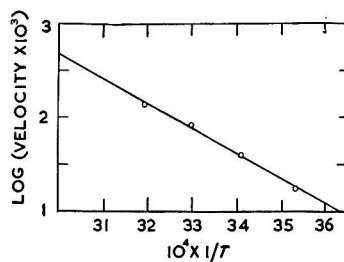


FIG. 4.—Arrhenius plot of the activity of sorghum β -amylase at various temperatures

Michaelis constant.—The Michaelis constant K_m was determined from a plot of initial velocity versus the ratio, velocity/starch concentration, as recommended by Hofstee¹⁸ (see Fig. 5). Two sets of results gave K_m values 0.77 and 0.78 g. starch/litre.

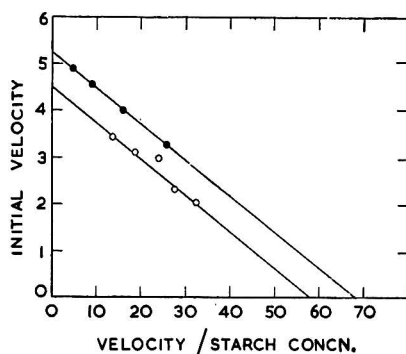


FIG. 5.—Determination of the K_m of sorghum β -amylase

Inhibition.—The β -amylases of barley malt, wheat and the sweet potato are known to be inactivated by copper and mercury compounds which combine with the essential sulphhydryl group,^{7, 8, 19} activity being restored by the addition of cysteine which removes the heavy metal. Sorghum β -amylase was also found to be very sensitive to inhibition by *p*-chloromercuribenzoate

(PCMB), one unit of amylase activity (K.D.U.) being 98% inhibited by 3.3×10^{-10} mole of PCMB. Practically complete reversal of inhibition was found on addition of cysteine. Typical data on the inhibition and its reversal are given in Table IV.

Table IV

Inhibition of sorghum β -amylase by p-chloromercuribenzoate (PCMB) and its reversal by addition of cysteine

PCMB, moles $\times 10^{-8}$	Amylase activity, K.D.U. after addition of		% Inhibition of original activity	% Recovery of original activity
	Inhibitor	Cysteine to inhibited solution		
0	227.6	—	0	—
0.5	169.3	225.4	25.6	99.5
1.25	115.1	236.4	49.4	103.9
2.5	48.4	218.6	78.7	96.1
5.0	18.2	225.8	92.0	99.2
7.5	4.7	192.8	97.9	84.7
10.0	0.0	—	100.0	—

Starch conversion limit.—The degree of starch conversion at pH 4.9 was 54.6% which is in fair agreement with the limit for barley β -amylase action, viz., $56 \pm 1\%$.²⁰

Conclusion

South African sorghum malts, unlike the American,¹ contain significant amounts of β -amylase, which is absent from the grain and is formed during germination at the same rate as the α -amylase so that the ratio of β - to α -amylase remains constant.

The properties of purified sorghum β -amylase closely resemble those of the other cereal β -amylases. The low diastatic power of kaffircorn malts compared with those of barley and wheat malts must be attributed therefore entirely to the quantitative difference in amylase composition, i.e., to kaffircorn malts possessing a far smaller proportion of β -amylase than do the malts of wheat and barley.

Acknowledgments

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KAFFIRCORN MALTING AND BREWING STUDIES. VI.*—Starch Content of Kaffir Beer Brewing Materials

By M. M. von HOLDT and J. C. BRAND

Starch was determined in kaffir beer brewing materials by a modification of the method of MacWilliam *et al.* Kaffircorn grains contained 61.1–69% starch; kaffircorn malts 45.9–61.2% and maize grits and maize meals 71.8–81.7%. The starch content of the spent grains from different breweries varied widely, from 14.7 to 50.3%. In a trial brew, 40.7% of the starch in the raw materials escaped degradation during mashing, 32.8% appearing in the final beer and 7.9% in the spent grains.

Introduction

A study of the activity of the amylases of kaffircorn malt during the brewing of kaffir beer¹ has indicated that mashing conditions in many breweries are such that the starch in the raw materials, particularly the malt, can be broken down only to a limited extent. It was therefore considered of importance to study the utilisation of starch in the brewing process in some detail. As a beginning, a survey has been made of the starch content of the raw materials used and of the spent grains or 'beer waste'.

Kaffir beer is brewed from kaffircorn malt, kaffircorn grain and various maize products.² Substantial data have been supplied on the starch content of grain sorghums grown in the United States of America,³ many of which originally came from South Africa, but there were no available data on the varieties grown in South Africa. Also, there was no information on the starch content as opposed to 'carbohydrate by difference' of kaffircorn malts and maize products of South Africa.

Since it was planned at a later stage to follow the breakdown of starch during malting and brewing, it was desirable to use a method for determining starch which was both specific as well as accurate. Many methods have been proposed for determining starch in cereals and cereal products and these have been reviewed by MacWilliam *et al.*,⁴ Radley⁵ and Herd & Kent-Jones.⁶ The method developed by MacWilliam *et al.*⁴ was selected as the most suitable for the present work. This method involves the extraction of the starch with perchloric acid, precipitation with iodine and final estimation of the carbohydrate with anthrone. The perchloric acid extraction, although satisfactory for barley grain and malt, failed to give complete extraction of the starch of kaffircorn grain and, therefore, some modification of the extraction procedure was necessary. Other modifications were introduced to increase the accuracy of the method and to render the method applicable to the analysis of small samples of malt produced in laboratory malting tests.

Experimental

All samples were ground to pass an 80-mesh sieve prior to analysis. The beer samples were centrifuged and the separated solids were dried at 50° and analysed for starch.

* Part V: preceding paper

Moisture was determined by heating the ground sample at 100° for 4 h. All results are expressed on a moisture-free basis.

Determination of starch

The following procedure was finally adopted.

(i) *Removal of the free sugars and water-soluble polysaccharides.*—Undried material (200 mg.) contained in a centrifuge tube (100 ml. capacity) was extracted under reflux on a boiling water-bath with three 10-ml. portions of 80% aqueous ethanol, each for 1 h., and the supernatants discarded. The residue, after being washed twice with water, was extracted three times with 5-ml. portions of water at 40° for 1 h. with stirring, and the supernatants again discarded.

(ii) *Dispersion of the starch in KOH and extraction with perchloric acid.*—Sand (200 mg.) and water (4.5 ml.) were added to the sugar-free material and the mixture cooled to 3°. 10N-KOH (0.5 ml.) was added with stirring, which was continued for 10 min. Water (5 ml.) was then added and stirring continued for a further 5 min. Four perchloric acid extractions were then carried out according to the method of MacWilliam *et al.*⁴ except that in the first extraction 8 ml. of perchloric acid were added instead of 6 ml.

(iii) *Precipitation with iodine.*—The procedure of MacWilliam *et al.* was followed, but, as centrifugation of the starch-iodine precipitate proved ineffective, the precipitate was filtered on to a pad of Celite contained in a sintered glass crucible (porosity 4) where it was washed and decomposed.

(iv) *Acid hydrolysis of starch and subsequent estimation of the resultant glucose.*—The starch-Celite mixture was transferred by means of a spatula into a hydrolysis tube. After addition of water (4.5 ml.) the tube was immersed in ice-water and thoroughly cooled. 10N-KOH (0.5 ml.) was added and the mixture stirred for 5 min. Water (5 ml.) was added and the stirring continued for 3 min. The solubilised starch was hydrolysed by adding 2.5N-H₂SO₄ (10 ml.) to bring the acid normality of the mixture to 1, sealing the tube and heating for 6 h. at 100°. The hydrolysate was then neutralised with NaOH (phenolphthalein), filtered into a 100-ml. volumetric flask, the paper thoroughly washed with water and the solution made up to volume. The concentration of glucose in aliquot portions (2 ml.) was determined by Somogyi's micro-method,⁷ and the corresponding weight of starch was obtained by multiplying the weight of glucose by 0.90.

Results and discussion

MacWilliam *et al.* removed the sugars and water-soluble polysaccharides from a large sample of material and carried out subsequent operations on aliquot portions of this. To make the method applicable to the small amounts of malt obtained in laboratory malting trials, the extraction was carried out on the weight of material required for the final determination, which had the advantage of reducing the time taken to remove the soluble carbohydrates.

The first starch determinations were carried out on the short red variety of kaffircorn. It was found that two perchloric acid extractions, as used in the analysis of barley and barley malt, were insufficient for complete removal of the starch and that four extractions were necessary. Similar results were obtained with all types of kaffircorn malt. Even more difficulty was encountered in extracting starch from the grain of certain other varieties of kaffircorn including white, Martin, Framida and birdproof. In all these cases starch was still present in the residue after seven perchloric acid extractions. Comparison of the amount of starch extracted by four perchloric acid treatments with that estimated to be present in the grain by the A.O.A.C. method⁸ showed that, while the two methods gave reasonably concordant results in the case of short red kaffircorn, the former gave results that were 8-11% lower than the latter in the case of Framida, Martin and white kaffircorn (Table I). While the A.O.A.C. method does not distinguish between starch and dextrans it is unlikely that mature grain will contain more than traces of the latter. Hence it appears that perchloric acid fails to extract a considerable proportion of the starch present in many varieties of kaffircorn.

Difficulties in extracting starch from sorghum have been reported by Watson *et al.*,⁹ who considered it to be due to the occurrence in the grain of a large proportion of horny endosperm

on which the starch granules are embedded in a thicker protein matrix than on the flouy endosperm. Difficulties in extracting starch from certain varieties of peas have also been reported to be due to association of the starch with protein.¹⁰ Attempts were therefore made to release the starch in the kaffircorn grain by proteinase action prior to perchloric acid extraction. White kaffircorn grain was treated with papain (0.2 and 5%) for 15 h. at 30°, but, although this procedure releases bound amylases of barley and kaffircorn,¹¹ it failed to facilitate extraction of starch. Attempts to use trypsin had to be abandoned as the enzyme preparation used apparently contained an amylase which completely degraded the starch.

Potter *et al.*¹² overcame similar difficulties experienced in extracting starch for fractionation studies by the use of N-KOH. When this treatment instead of water gelatinisation was applied to kaffircorn quantitative extraction of starch was obtained from all varieties of grain and brewing materials with 4 perchloric acid extractions (cf. Table I). This procedure was therefore adopted as standard.

Table I

Comparison of the starch content of kaffircorn grains by the A.O.A.C. method and by perchloric acid extraction with and without prior gelatinisation with KOH

Variety of grain	Starch content (% dry weight)		
	A.O.A.C. Method	HClO ₄ extraction	
		Without pretreatment	With pretreatment
Short red	65.3	69.9	69.9
Framida	59.6	50.0	61.1
Martin	63.5	52.6	63.2
White	63.0	55.1	64.5

MacWilliam *et al.* favoured the estimation of the extracted starch with anthrone-sulphuric acid, since they found that dilute acid hydrolysis of the starch resulted in only 96–97% recovery of the glucose residues. Our experience, however, has shown the anthrone method to be far less reliable than hydrolysis of the starch followed by estimation of the resultant glucose by Somogyi's micro-method. If hydrolysis is carried out in a sealed tube for an optimum period, degradation is at a minimum and 98% of the glucose can be recovered. It was also found that solubilisation of the starch prior to acid hydrolysis with KOH in the cold instead of with NaOH at 100° resulted in improved recovery.

The method finally adopted gave an overall recovery of 97.7% on samples of pure sorghum starch.

Starch content of brewing materials

The starch contents of a variety of raw materials used in kaffir beer brewing are shown in Table II. The values obtained for the kaffircorn grain samples are in the same range as those obtained by Horan & Heider³ for grain sorghums in the United States. The kaffircorn malts examined here were all commercial products. As expected, their starch contents were considerably lower than those of the grains. A detailed study of the change in starch content during malting has been made and is reported in the following paper.¹³

The maize products most commonly used in kaffir beer brewing are maize grits and maize meal. The former is a crushed degermed meal, whereas the latter is essentially ground whole maize, the various subgrades of which differ only in the degree of fineness of the product. As will be seen from Table II, these products have a higher starch content than has kaffircorn grain. Since they are also cheaper, there is an increasing tendency for breweries to switch over from kaffircorn grain to maize meal or grits as a brewing adjunct.

As will be seen from Table III, the starch content of the spent grains from different breweries varies considerably, from 14.7 to 50.3%. The beer waste with the highest starch content was from a brewery which used a brewing recipe with a very high proportion of malt to unmalted grain.

The results of an attempt to follow the fate of the starch in brewing are shown in Table IV. From this it will be seen that 40.7% of the starch was not broken down during mashing. Of

this, 7.9% was discarded in the spent grains, while 32.8% appeared in the finished beer. The presence of this starch, which amounted to 3.6% by weight in the beer examined, gives kaffir beer a considerable viscosity, and is largely responsible for the 'creminess' of texture which is an important factor in consumer acceptance. It also helps to prevent the insoluble solids in the beer from settling out on keeping. Further studies of the breakdown of starch during the brewing process and its influence on beer quality will be reported in a later communication.

Table II

Starch content of raw materials used in kaffir beer brewing

Material	Starch content (% dry basis)	Material	Starch content (% dry basis)	Material	Starch content (% dry basis)
<i>Kaffircorn grains</i>		<i>Kaffircorn malts</i>		<i>Maize products</i>	
Short red	69.0	Short red 1	55.5	Maize grits 1	81.7
Framida	61.1	" " 2	46.7	" " 2	75.3
Martin	63.2	" " 3	47.2	Unsifted, ungranulated maize meal	71.8
White	64.5	" " 4	45.9	Unsifted, granulated maize meal 1	72.1
Birdproof	68.6	" " 5	61.2	" " " " 2	72.1
		Birdproof 1	55.0	Sifted granulated maize meal	78.0
		" 2	53.8		
		" 3	52.4		
		Martin	51.0		

Table III

Starch content of spent grains

Origin	% Starch
Brewery A	36.2
" B	14.7
" C	50.3
" D	30.1

Table IV

Fate of starch during brewing

Raw materials	Quantity	Starch content %	Weight of starch, lb.	% of total starch
Kaffircorn malt	802 lb.	61.2	491	29.1
Maize grits	1591 lb.	75.3	1199	70.9
				100.0
Products				
Beer	1530 gal.	3.6	554	32.8
Spent grains	445 lb.	30.1	134	7.9
				40.7

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**KAFFIRCORN MALTING AND BREWING STUDIES. VII.*—
Changes in the Carbohydrates of Kaffircorn during Malting**

By M. M. von HOLDT and J. C. BRAND

The sugars present in the kaffircorn grain and malt have been identified: glucose, fructose and sucrose are present in the grain, while the malt contains also maltose and lower maltose oligosaccharides. Data are given for the changes occurring in the content of these sugars and starch during germination of the kaffircorn and drying of the malt at 50°.

Introduction

The malting of kaffircorn in South Africa¹ is carried out under conditions which differ considerably from those used for barley. Kaffircorn is usually steeped for 8–16 h., which is very short by barley standards, and the grain is allowed to germinate at a very much higher temperature than is barley. In the outdoor floor maltings, temperature control is very crude and the temperature of the bed varies between wide limits depending on the season of the year. In the pneumatic maltings, however, it is attempted to keep the germinating grain between 25 and 30°, which has been shown in this laboratory to be optimum for diastatic power development.² The grain is watered daily during the first part of the period of growth, which is generally 5–7 days. Under these conditions a much greater development of roots and shoots occurs than in barley malt. Kaffircorn is not kilned in the sense understood by the barley malt industry but is dried at a temperature not exceeding 50° to minimise destruction of the amylase activity.

A number of qualitative and quantitative studies have been made of the changes taking place in the carbohydrates of barley during malting,^{3–5} but no comparable data were available for kaffircorn malts. It was therefore considered to be of interest to study the changes in the content of starch and simpler sugars in a typical kaffircorn when malted in the laboratory under conditions giving maximum diastatic power development.

Experimental

Materials

The short red variety of kaffircorn was used for these studies. This variety is typical of those which comprise the K2 Red grade of kaffircorn which is most commonly used for malt manufacture.

Preparation of malts

The samples for the examination of the sugars were prepared as follows. After being steeped for 8 h. at 20–25°, the grain was germinated at 30° for 7 days. Two samples were collected on each day except the 6th. One was dried in a forced-draught oven for 24 h. at 50°,

* Part VI: preceding paper

which is the temperature normally used for drying kaffircorn malt in this laboratory (sample D). In order to observe whether the heating at 50° affected the sugars in any way, the other sample was dropped into boiling absolute alcohol and refluxed for 30 min. to inactivate the enzymes. The alcohol was then filtered off and the remaining corns, roots and shoots were dried and milled. The filtrate was then concentrated to dryness and the residue thus obtained was combined with the milled product (sample W).

The samples for the investigation of the changes in starch content during malting were prepared 2 years after the first series under essentially the same conditions. A sample was collected on each day, except the first, and dried for 24 h. at 50°. The roots and shoots were removed before analysis of the corns.

Analytical methods

Samples were ground to pass a 60-mesh sieve.

Moisture determinations were carried out *in vacuo* at 40°.

Diastatic power was determined by the aqueous extraction method of Novellie.⁶

Starch was determined as described by von Holdt & Brand.⁷

Sugars.—Concentration of solutions was carried out at 40°/20 mm.

(a) *Qualitative identification*.—A sample of a 7-day malt (350 g.) was boiled with absolute ethanol on a water-bath for 30 min. to inactivate the enzymes. After filtration, the sugars were extracted by macerating the malt in a Waring Blendor with 80% aqueous ethanol (1 l.). After centrifugation, two further extractions were carried out by shaking the residue with 80% ethanol (1 l.) at room temperature for 3 h. All the alcoholic extracts were combined and concentrated to a small volume, de-ionised with Amberlite IR-120 and IR-4B resins and finally concentrated to a syrup (18 g.), which was adsorbed on to a cellulose column (42 × 7½ cm.). Elution was carried out with butanol-water (9 : 1), followed by butanol-ethanol-water (3 : 1 : 1) and then butanol-ethanol-water (2 : 2 : 1). Six fractions were thus obtained :

Fractions (1)–(4) were recrystallised and identified by mixed melting point and comparison of the optical rotation with that of the authentic sugar. Fractions (1)–(4) were thus found to be D-fructose (0.8 g.), D-glucose (5.6 g.), sucrose (3.6 g.), and maltose (1.6 g.), respectively.

Fraction (5) (0.8 g.) failed to crystallise as did its acetate. On hydrolysis it gave rise to glucose only and estimation of the number of sugar units by the method of Whelan *et al.*⁸ showed it to be a disaccharide. Periodate oxidation⁹ produced no formaldehyde, thus indicating the presence of a 1,6-linkage in the molecule. Although not conclusively proved, the sugar appeared to be isomaltose.

Fraction (6) (1.1 g.) failed to crystallise $[\alpha]_D^{25} + 151.3^\circ$ (c, 1.2 in water). Hydrolysis yielded glucose only and estimation of the number of glucose units showed it to be a trisaccharide indicating that the original sugar was probably maltotriose.

The free sugars were extracted from the kaffircorn grain (1 kg.) and separated on a cellulose column in exactly the same manner as for the malt. Three fractions were thus obtained which were identical with fractions (1), (2) and (3) of the malt.

(b) *Quantitative estimation of the free sugars*.—The sugars were extracted essentially according to the method of MacLeod.¹⁰ After inactivation of the enzymes with absolute ethanol (125 ml.), the grain (50 g.) or malt (10 g.) was macerated in a Waring Blendor for 5 min. with 80% aqueous ethanol and the mixture then centrifuged. The sugars were extracted from the residue by shaking with 80% ethanol (150 ml.) at room temperature for 1 h. Three such extractions were necessary for complete removal of the sugars. A weighed amount of rhamnose, which would later act as a reference sugar during quantitative chromatography, was added to the combined alcoholic extracts. After concentration to a small volume, the solution was de-ionised with Amberlite IR-120 and IR-4B resins and again concentrated in preparation for spotting on paper. (De-ionisation was essential for a satisfactory separation of the sugars.) The concentrate was spotted on Whatman's No. 1 chromatographic paper and developed in ethyl acetate-acetic acid-formic acid-water (18 : 3 : 1 : 4). The positions of the sugars on the paper were located in the usual way by means of marker strips. Each strip of paper containing

a sugar was then cut out, macerated with water (20 ml.) and the mixture filtered. The sugars in the filtrates (2 ml. aliquots containing 0.03-0.06 mg. of monosaccharides and 0.05-0.10 of lower oligosaccharides) were estimated by Somogyi's micro-method.¹¹

Results

All results are expressed on a moisture-free basis. Results for the second and first maltings are shown in Tables I and II, respectively, and the changes in the free sugar content of the samples are summarised in Table III on a weight basis and Fig. 1 on a 1000-corn basis.

Table I

Changes in the starch content of short red kaffircorn during malting

Sample	Days of germination	Diastatic power, K.D.U./g.	1000 Corn wt.		Starch, g./100 g.	Starch, g./1000 corns
			Whole malt	Corns alone*		
Grain	0	0	—	24.9	71.0	17.7
Malt 2	2	26.6	23.5	22.7	68.7	16.2
"	3	56.3	22.9	21.0	64.9	14.8
"	4	63.7	22.3	20.0	63.8	14.2
"	5	68.5	21.4	18.7	59.9	12.8
"	6	60.5	20.3	17.3	56.5	11.5
"	7	45.9	18.8	15.3	53.5	10.0

* Minus roots and shoots

Table II

1000 Corn weight and diastatic power of samples in first malting

Sample	Days of germination	Diastatic power, K.D.U./g.	1000 Corn wt. (g.) (Whole malt)
Grain	0	Nil	
Malt 1	1	11.5	27.2
"	2	30.5	27.1
"	3	41.3	26.1
"	4	54.9	24.7
"	5	59.1	23.9
"	7	82.8	21.3

Table III

Changes in the free sugars of short red kaffircorn during malting

Sample	Days of germination	Sugar, g./100 g. of malt							
		Fructose		Glucose		Sucrose		Maltose and maltose oligosaccharides	
Grain	0	0.03		0.04		0.89		Nil	
		W*	D*	W	D	W	D	W	D
Malt 1	1	0.25	0.20	0.50	0.54	1.34	1.51	0.19	<0.1
"	2	0.55	0.48	1.45	1.48	2.32	3.07	0.90	0.53
"	3	0.77	0.62	1.57	1.85	2.79	3.89	3.25	0.94
"	4	1.23	0.97	2.39	2.91	3.86	4.61	3.72	1.40
"	5	1.73	1.31	3.47	4.14	4.08	5.19	4.24	2.44
"	7	1.90	1.57	6.32	5.48	4.30	5.58	8.05	3.33

* W = sample dropped into boiling alcohol—see Experimental part
 D = sample dried at 50°

Discussion and conclusions

During the malting of kaffircorn, the starch content of the corns decreased by 43% from 17.7 to 10.1 g./1000 corns (Table I). The extent of starch breakdown is thus considerably greater than is encountered in barley malting where Hall *et al.*⁵ reported a decrease of 20%.

This is not surprising in view of the higher temperatures used in the malting of kaffircorn and the more vigorous growth of the malt.

Glucose, fructose and sucrose are the only sugars present in ungerminated kaffircorn grain (Table III). MacLeod *et al.*³ and Harris & MacWilliam⁴ found gluco-difuctose, raffinose and a trace of maltose in addition to the above sugars in ungerminated barley grain. This fits in with de Cugnac's^{1,2} classification of grasses into two distinct types: (i) those which form fructosans, e.g., wheat, barley, oats and rye; (ii) those which do not form fructosans, e.g., maize, sugar cane, rice and sorghum. In both the kaffircorn and the barley grain, sucrose was the predominating sugar.

The germination of kaffircorn was accompanied by a steady increase of fructose and sucrose which was apparent from the first day and continued until the fifth day, after which there was virtually no change (Table III and Fig. 1). Maltose, isomaltose, maltotriose and traces of higher maltose oligosaccharides appeared on the first day and increased sharply over the whole 7-day period, as did glucose.

Unfortunately it was not possible to follow the changes in the starch and sugars in the same series of malts and so to determine the net carbohydrate balance during malting. Loss of starch, which was equivalent to a loss of 30% of the grain was, however, very much greater than the increase in the content of sugars and lower oligosaccharides, equivalent to 14.7% of the original weight of the grain (calculated as g./100 g. of original grain) in the first series. It thus appears that there is a considerable net loss of carbohydrate in the malting of kaffircorn.

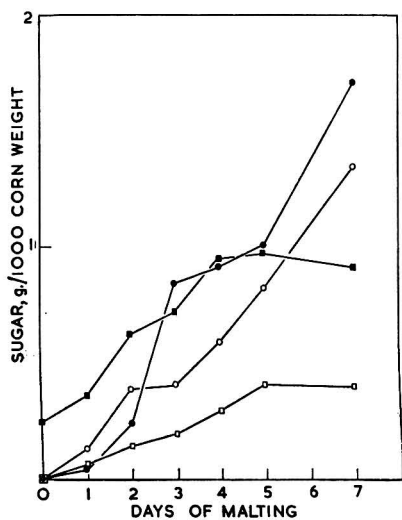


FIG. 1.—Changes in the free sugars of short red kaffircorn during malting

Fructose □
 Glucose ○
 Sucrose ■
 Maltose plus maltose oligosaccharides ●

The pattern of the development of sugars in the malting of kaffircorn is similar to that found in barley. The main differences can be traced to the more rapid growth of the former grains. Thus in barley MacLeod *et al.*³ and Hall *et al.*⁵ found virtually no change in the content of glucose, fructose and maltose during the first 4 days of germination, while the sucrose content decreased. MacLeod postulated that in the early stages of germination sucrose was providing material for respiration, synthesis or both and sucrose production was outstripped by consumption. Only after degradation of starch and fructosans had commenced did the sucrose increase. As pointed out above, the breakdown of starch during germination of kaffircorn is much more rapid, hence the production of sugars probably always exceeded the consumption, so that there was no lag period before the sugar content of the grain started to increase.

Comparison of the sugar content of the malt dried at 50° with that of the alcohol-treated samples (Table III) shows that heating at 50° resulted in the disappearance of approximately 50% of the maltose and maltose oligosaccharides. The fructose content of the malt also decreased, but to a somewhat lesser extent. On the other hand, there was a significant increase in

the sucrose content. The glucose content remained more or less the same. Maillard-type reactions probably play a part in the decrease of maltose and fructose through breakdown of the maltose and maltose oligosaccharides to glucose, and synthesis of sucrose from fructose may also be involved, as has been postulated in the kilning of barley malt.^{3, 4}

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SELECTION OF A PYRETHRUM-RESISTANT STRAIN OF THE GRAIN WEEVIL, *CALANDRA GRANARIA* L.*

By E. A. PARKIN and C. J. LLOYD

Adults of a field strain of the grain weevil, *Calandra granaria* L., initially twice as resistant to pyrethrins in oil solution as a laboratory standard strain, have been exposed to selection pressure with pyrethrins in 17 out of 22 generations in the course of five years. The strain has steadily increased in resistance to pyrethrins and is now 18 times as resistant as the standard. There has been a simultaneous increase in resistance to pyrethrins synergised with piperonyl butoxide of only $\times 2$.

In the course of selection the beetles of the resistant strain became heavier in weight but not sufficiently to account for the resistance. They also became darker in colour but this change was shown to be physiologically unconnected with the increase in resistance to pyrethrins.

Introduction

The induction of resistance to insecticides in strains of insects of agricultural, and especially of public health, importance is now well established. There would seem to be no reason why

* Read at the Pyrethrum Symposium of the Pesticides Group, 16th November, 1959

similarly resistant strains should not be expected among insects infesting stored foodstuffs but their development is likely to be slow because of the artificial changes of population that occur and the relatively long length of life-cycle of most of the species concerned. This latter factor has doubtless also discouraged attempts to select resistant strains under laboratory conditions of exposure.

Records of resistance in stored-product insects are relatively few and all but perhaps one refer to the laboratory selection of strains that appear to fall in the category showing 'vigour tolerance' as defined by Hoskins & Gordon,¹ bearing in mind that any resistance factor that is determined is partly a function of the test method used. Mathlein² increased the resistance of *Calandra granaria* slightly through selecting the adults for seven generations by exposure to DDT dust. Another attempt to induce resistance to DDT was made by Maeda,³ who bred *Tribolium confusum* in flour containing various concentrations of DDT and evoked increases of resistance from $\times 2$ to $\times 8$ in 10–15 generations after which the resistance level stabilised.

Exposure of *C. granaria* weevils to filter-papers treated with pyrethrins and breeding from the survivors after 85% knock-down led to no increase in resistance for six generations, then a rise to $\times 3\frac{1}{2}$ by the tenth generation, with no further change up to the fifteenth generation inclusive (Blackith⁴). The results of a comparison by Holborn⁵ of resistance to kill by a pyrethrin dust mixed with grain between a laboratory strain of *C. granaria* and a strain obtained from a treated farm granary showed that the latter strain was $3\frac{1}{2}$ – $6\frac{1}{2}$ times as resistant as the former. It was the provision of a supply of beetles from this slightly resistant granary strain that led to the start of the present investigation to determine whether the resistance of a strain of a stored-product insect to a contact insecticide could be raised clearly beyond the level of possible confusion with vigour tolerance.

More prolonged efforts to induce resistance have been made with fumigants. Monro & Uptis⁶ selected a laboratory and a 'wild' strain of *C. granaria* by exposing each generation of adults to methyl bromide. In the course of 14 generations both selected strains only doubled their LD₅₀ values. Hope (unpublished) has applied selection pressure to the Pest Infestation Laboratory strain of *Tribolium confusum* with methyl formate for 35 generations and achieved an increase in resistance to this compound of about $\times 3$. It is clear that in neither instance has an outstanding level of resistance been effected.

Against all these deliberate attempts to induce resistance must be set the experience of Blackith & Gorringer⁷ who accidentally increased the resistance of the eggs of a strain of *Calandra granaria* to mercury vapour in less than 10 generations by the extraordinary factor of at least 350, calculated on the basis of concentration \times time products.

Experimental

Origin of the resistant strain

In September, 1954, the Cooper Technical Bureau supplied us with a culture of a strain of *Calandra granaria* L. which they had found to be slightly resistant to pyrethrins. According to the Bureau, the strain was collected in October, 1953, from a farm granary in which, during the previous 3 years, a few small-scale experiments had been undertaken entailing the admixture with bagged wheat of grain-protectant dusts containing pyrethrins and piperonyl butoxide. Jute bags treated with oil solutions of the same insecticidal mixture had been used in other experiments. At no time had the experimental work involved more than a small fraction of the contents of the granary and no resistance was noted in the course of the tests.

When in February–April, 1954, the Cooper Technical Bureau compared the resistance of the granary strain with that of their current laboratory strain to the synergised pyrethrin powder, no marked difference in susceptibility of the adult weevils was found. When the strains were compared using a non-synergised pyrethrin powder, however, the granary strain was found to have an LC₅₀ between $3\frac{1}{2}$ and $6\frac{1}{2}$ times that of the current laboratory strain.⁵ Since then, the granary strain has been maintained in laboratory culture at the Bureau without any attempt to build up the resistance and, when re-tested with non-synergised pyrethrum powder in March, 1958, its resistance had fallen during the 4 years' interval to about twice that of their current laboratory strain. During the whole of this culturing period both strains were bred at 25° and 60% R.H. on wheat.

Laboratory culture of resistant strain

Since receipt at the Pest Infestation Laboratory (P.I.L.), the granary strain has been maintained in jar cultures at 25° and 70% R.H. at a general breeding rate of just less than five generations per annum. Each cycle has started with about 80 weevils placed on approximately 270 g. of sterilised Manitoba No. 2 wheat, the parent beetles being removed by sieving 4 weeks later. Since emergence of the offspring begins towards the end of the fifth week, sieving the culture in the seventh and ninth weeks has provided two batches of adults 0–2 weeks old. These have been kept in groups of 525 on about 135 g. of wheat for two weeks and then subjected to insecticidal treatment when 2–4 weeks old: supplementary tests have occasionally been made a week later with any remainder of beetles, which were then 3–5 weeks old.

The resistant strain was bred side by side with the standard strain of the Pest Infestation Laboratory, Insecticide Section, against which comparisons of resistance were made at intervals.

Insecticidal techniques

Selection pressure was maintained on the resistant strain by treating successive generations with pyrethrins and breeding from the survivors. Between 500 and 750 insects were treated in batches of 50 on each occasion and a mortality of about 80% was aimed at; the actual percentage kills varied considerably, although most fell between 60% and 90%. When sufficient adults were available, their resistance relative to the standard strain was determined. The same general types of procedure were used for both the selection experiments and the determination of relative resistance.

Initially, some tests were made with the film technique⁸ but it was considered advisable to make food available to the beetles and this was not possible without contamination from the films. The early experiments (generations F_1 to F_6) were therefore done with the direct spray technique,⁹ but a little broken wheat was provided as food 24 h. after the insects were treated. Solutions for spraying were made on a weight/volume basis by diluting with Shell Risella oil No. 17 (R17) a pyrethrin concentrate containing in the same oil 6.1% w/v total pyrethrins as determined by the A.O.A.C. method. The rise in resistance of the beetles necessitated increases in the concentration of pyrethrins applied with consequent increase in viscosity of the spray liquid and, ultimately, uneven distribution of the spray. Change to a 15% w/w pyrethrin concentrate in F_6 gave only a brief relief and in F_{10} the direct spray technique was abandoned in favour of individual topical treatment of beetles. They were anaesthetised with carbon dioxide and dosed by means of a micro-capillary tube.¹⁰

In the topical application treatments, the 15% pyrethrin concentrate was diluted with purified cyclohexanone (containing about 2% of cyclohexanol) but after F_{17} up to 50% of the cyclohexanone was replaced with light petroleum (b.p. 100–120° C) in order to reduce the viscosity further during dosage. A standard volume of 0.03 μ l. was applied to the abdominal sternites of each beetle. This dosage of cyclohexanone alone had no detectable effect on the insects. After treatment the insects were confined on filter papers within glass rings and a small quantity of broken wheat was added as food. In both direct spray and topical application tests up to F_{14} the results were computed on the basis of the reactions of the insects as observed on the sixth day of exposure but, from F_{15} onwards, the period was extended to 8–14 days so that the final effects of the treatment became evident in that all paralysed beetles had either recovered or succumbed. The criteria for paralysis and death were those given by Hewlett.⁹

In determinations of relative potency by the direct spray technique, two replicates of 50 beetles were treated at each of four concentrations; when this method was superseded by topical application, two replicates of 30 beetles were treated at each of three concentrations.

Selection experiments

The progress of the experiments in which selection pressure was applied is summarised in Table I. Where generations are not included in the table, they were bred through without selection because sufficient numbers of insects were not available, for example, in F_{11} as a result of a 98% kill in F_{10} , or in F_{16} when numbers of beetles were required for special tests.

Table I

Data showing how selection pressure has been applied to *C. granaria* to induce increase in resistance to pyrethrins

Generation	Type of test	Pyrethrins concn. % w/v	Deposit	Kill %
F ₂	Direct spray	1.2	4.5 mg./10 cm. ²	70
F ₃	" "	1.3	4.3 "	41
F ₄	" "	1.55	4.5 "	66
F ₅	" "	1.8	4.0 "	53
F ₆	" "	2.15	4.5 "	66
F ₇	" "	2.7	4.5 "	65
F ₉	" "	4.0	4.1 "	77
F ₁₀	Topical application	5.0	0.03 µl.	98
F ₁₂	" "	5.0	" "	87
F ₁₃	" "	4.91	" "	88
F ₁₄	" "	5.45	" "	87
F ₁₇	" "	6.04	" "	82
F ₁₈	" "	7.0	" "	70
F ₁₉	" "	7.5	" "	79
F ₂₀	" "	8.0	" "	65
F ₂₁	" "	8.75	" "	64
F ₂₂	" "	10.0	" "	77

Furthermore, F₀ and F₁ were not subjected to selection as the strain was then being established in our cultures and its resistance measured.

The results show that, as the generations proceed, the pyrethrin concentration used in applying the selection pressure had steadily to be increased from 1.2% to 10.0%. The actual deposit received by a beetle in the direct spray tests has been estimated from available data (Gostick, unpublished) as an average of 0.025 µl. of the oil solution, so that, apart from the change in method of application, virtually the same mean volume of insecticidal solution has been given to each beetle throughout.

Results

Change in resistance to pyrethrins

Measurements of the resistance of the selected strain in comparison with that of the standard strain were made in several of the generations. The experimental data were transformed to probit mortality and log concentrations and provisional regression lines were fitted by eye: it was thought unnecessary for the purposes of this investigation to derive relative potencies more precisely by computation. There was no indication of any marked or sustained departure from parallelism of the pairs of regression lines representing the reactions of the two strains. The results are summarised in Table II.

Table II

Comparisons of the resistance to pyrethrins of the selected strain (R) and the standard strain (S) of *C. granaria*

Generation	Type of test	LC ₅₀ of S strain (% w/v)	Relative resistance R : S
F ₁	Direct spray	0.60	2 : 1
F ₈	" "	0.48	5 : 1
F ₁₇	Topical application	0.40	12 : 1
F ₁₈	" "	0.31	16 : 1
F ₁₉	" "	0.47	14 : 1
F ₂₁	" "	0.43	18 : 1

The possible effect of the change in technique of application of the insecticide is not known but, because of the basic similarities of the two techniques and the approximate equality of the volumes of solution applied to each insect, no major difference would be expected. There was some variability in the resistance of the standard strain as shown by the concentrations of pyrethrins needed to cause 50% kill. The data of Table II, however, in no way contradict

the conclusion that there has been a very marked increase in the resistance to pyrethrins of the selected strain.

Three tests by the film technique made on F_0 , F_1 and F_8 respectively did not give acceptable results because of high mortality of the controls or the need to extrapolate unduly beyond the results actually obtained in the experiments. It appeared, however, that the resistance factor between the two strains was likely to be greater by the film technique than by direct spray or topical application.

Resistance to pyrethrins with piperonyl butoxide

It will be recalled that the only known original exposure of the selected strain was to a pyrethrins-piperonyl butoxide powder, and possibly to impregnated sacking, in the granary. The available adults of F_{16} were therefore used to test the reaction of the pyrethrin-selected strain to the synergistic mixture. Preliminary tests were made to determine the range of concentrations to be used in the topical application of solutions containing pyrethrins and piperonyl butoxide in the constant ratio 1:10. The main experiments then showed that with the standard strain the LC_{50} for pyrethrins in R17 oil (with cyclohexanone) was 0.35% w/v, but was only 0.076% when the synergist was included; this gives a ratio of 4.6:1 between the concentrations of non-synergised and synergised pyrethrins needed for equal kill. The corresponding values with the selected strain were 5.5% for pyrethrins alone, 0.15% pyrethrins in the synergistic mixture—a ratio of 37:1. Whereas the ratio of the LC_{50} between these two strains for pyrethrins alone is $5.5/0.35 = 15.7:1$, the ratio for the synergised pyrethrins is $0.15/0.076 = 2.0:1$. From Table II it might be expected that the relative resistance of the selected strain in F_{16} would be somewhat less than 15.7:1, but the two strains had to be tested at separate times and not simultaneously as in those tests included in the table.

It is clear that selection of the strain for resistance to pyrethrins alone has not involved an equal resistance to pyrethrins synergised by piperonyl butoxide.

Change in size of the selected strain

On receipt of the granary strain from the Cooper Technical Bureau, the adults appeared to be very slightly smaller than those of the P.I.L. standard strain, even after breeding under the same general conditions for one generation: this suspicion was confirmed by weighing. In the course of the pyrethrin-selection experiments, the granary strain increased in size and has now (F_{22}) become distinctly larger than the standard strain. This change is reflected in the weights recorded in Table III.

Table III

Change in weight of pyrethrum-resistant and standard strains

Date	Generation	Wt. of 100 beetles, g.		Ratio R:S
		Resistant	Standard	
3.1.55	F_1	0.2336	0.2752	0.85:1
9.11.56	F_{10}	0.2705	0.2532	1.07:1
15.5.59	F_{22}	0.3446	0.2738	1.26:1

During the experimental period of over 4 years the selected strain has increased in average weight by nearly 50%, without a corresponding gain by the standard strain. The reason for and significance of this increase in weight are not known but the gain in weight is clearly insufficient to account for the increase in resistance to pyrethrins of the R strain.

Change in colour of the selected strain

When it was received, the granary strain appeared to be slightly darker brown in colour than the P.I.L. standard strain and it has become progressively darker in colour as the selective breeding for resistance to pyrethrins has continued. The range of colour in the standard strain is narrow, but in the resistant strain is wide because of the presence of much darker forms. In the tenth generation of the selected strain, groups of the palest and darkest individuals were picked out and cultured separately for five more generations, selection for colour being repeated in each generation. The two lots of eventual progeny which, *en masse*, were easily distinguishable by colour, were topically treated with three concentrations of pyrethrins in

the range 1.23–2.43%. Computation of the regression lines showed that the pale beetles were 1.2 times as resistant as the dark and that this difference was highly significant ($P = 0.01$). It may therefore be concluded that the darkness of colour is not physiologically connected with the increase in resistance to pyrethrins. A tendency for insecticidal selection to produce darker forms has nevertheless been noted by other workers.^{11, 12}

Discussion

It is interesting to note that the exposure of the original strain of *Calandra granaria* in the granary to insecticidal treatment involving pyrethrins synergised with piperonyl butoxide was not at all severe, being very local and of relatively short duration. It was not such, in fact, as would lead to the expectation of resistance. It is equally interesting that Holborn's initial laboratory tests showed a significant increase in resistance to pyrethrins but not to the synergised mixture that may have caused the change.⁵ Submission of the strain to laboratory selection with pyrethrins alone has evoked a fairly steady rate of increase in the resistance of the beetles to this insecticide. Although 22 generations of the insect have so far been reared, experimental difficulties made the application of selection pressure possible in only 17 generations and in some of these the pressure could, with advantage perhaps, have been somewhat higher. Nevertheless, this series of selections has raised the factor of resistance to pyrethrins under our experimental conditions from an initial $\times 2$ to the present $\times 18$. This is the greatest increase in resistance to any insecticide so far recorded among stored-product insects.

Apart from the two references mentioned earlier of induced resistance of stored-product insects to pyrethrins,^{4, 5} records of resistance to this insecticide among other insects are concerned with houseflies, mosquitos or cockroaches which were either selected in the laboratory for resistance or, more frequently, were strains exhibiting cross-tolerance to pyrethrins at the level of vigour tolerance. Among the former, Brown¹³ quotes Chadwick who raised the resistance of a laboratory strain of houseflies only 6–10 times by selection over 220 generations. Increase in resistance of houseflies to pyrethrins synergised with piperonyl butoxide has been induced in the laboratory by Decker & Bruce,¹⁴ who raised the resistance factor 10–20 times in 30–40 generations; and by Davies *et al.*,¹⁵ who found a rapid increase of $\times 11$ – 14 as a result of practical application of the synergised insecticide in powder form on farms in Sweden. Busvine¹⁶ reported receiving an Italian strain of DDT-resistant housefly five times as resistant to pyrethrins as another DDT-susceptible strain; on selection for pyrethrin resistance in seven of the next twelve generations, the resistance increased to only 6.5.

A field strain of the cockroach, *Blattella germanica*, showing resistance to chlordane, dieldrin and lindane was found to be 13 times as resistant to pyrethrins as a 'normal' laboratory strain and 29 times as resistant as the National Pest Control Association's strain.¹⁷ A high level of cross-tolerance to pyrethrins, although not in an insect, has recently been reported from South Africa by Whitehead¹⁸ for a field strain of the Blue tick, *Boophilus decoloratus*, resistant to sodium arsenite, BHC and DDT. This strain was 18.1 times more resistant to pyrethrins than his susceptible strain, although pyrethrins have apparently never been used in practice in that country for the control of ticks. Strains resistant to BHC or to sodium arsenite + BHC were only 2.4 times as resistant to pyrethrins.

The general picture is, therefore, that marked resistance to pyrethrins does not develop readily in the course of laboratory selective breeding but, in suitable circumstances, may appear more quickly as a consequence of application of synergised or straight pyrethrins in practice. With so expensive an insecticide as pyrethrum, however, a relatively small increase in resistance may well be sufficient to make its use no longer economic.

As its resistance is still increasing, the P.I.L. pyrethrin-resistant strain must be genetically heterozygous and no forecast can be made of how high the resistance may ultimately rise. If it rises much more, further experimental difficulties may be encountered in applying sufficient quantities of the pyrethrins to maintain the selection pressure. However, the enforced modifications of technique already made have not markedly varied the rate of application of the insecticidal solution and any changes in insecticidal toxicity resulting from alteration of the viscosity of the solutions have apparently been of minor importance compared with the increase in resistance.

Working with strains of *T. castaneum* of relatively low resistance to pyrethrins, Holborn⁵ hinted that piperonyl butoxide synergised pyrethrins better against the more resistant strains. The difference in resistance of the P.I.L. resistant strain to pyrethrins and to the synergised mixture is most marked. However, when the resistance of insects is increased by selective exposure to pyrethrins synergised with piperonyl butoxide, this difference may not occur as evidenced by the Swedish fly strain exposed on farms to a synergised pyrethrum powder and subsequently found in laboratory tests to have approximately the same resistance factor to pyrethrins alone as to the synergistic mixture.¹⁵

The existence of highly resistant and non-resistant strains of the grain weevil now offers possibilities of extending our knowledge of the mode of action of the pyrethrins and of the mechanism of their synergism, notably with the methylenedioxyphenyl compounds. Other points also await investigation such as the resistance of this strain to related insecticides like allethrin, to unrelated insecticides and to environmental stress.

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RENDZINAS AND RED-BROWN SOILS ON LIMESTONE: GENETIC INTER-RELATIONSHIP

By D. H. KHAN*

A study of the chemistry of rendzinas and red-brown soils and the insoluble silicate residue of the underlying limestone rocks was carried out to ascertain any genetic inter-relationship on the basis of Robinson's hypothetical conjecture. Rendzinas and red-brown soils are fairly correlated with the insoluble silicate residue of the corresponding underlying limestone rocks, and it is not necessary to postulate that the red-brown soils are formed by decalcification and desilicification of rendzinas.

Introduction

Robinson¹ has distinguished two main groups of soils derived from or associated with calcareous materials, such as limestone, chalk or coral: (i) rendzina soils of high base status, with a grey or greyish brown colour associated with a siliceous type of clay, probably devoid

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of free sesquioxides ; (ii) red and brown soils of low base status, with a red or reddish brown colour associated with clay having $\text{SiO}_2/\text{R}_2\text{O}_3$ ratio of about 2.0 or less, in which there is a certain proportion of sesquioxides.

Formation of the second group is, according to Robinson, due to decalcification followed by desilicification of the first group. This process would result in lowering of base status and $\text{SiO}_2/\text{R}_2\text{O}_3$ ratio, and an increase in free sesquioxides, especially iron oxides. This idea of a continuous soil-forming sequence, beginning from rendzina to red-brown soil, is also shared by Kubiena.²

Several other authors, notably Glinka,³ Nevros & Zvorykin⁴ and Stephens,^{5, 6} have a different view and have suggested that the red and brown soils occur mainly in association with hard crystalline limestones. Reifenberg⁷ has pointed out that the weathering of soft calcareous rocks seldom results in the formation of red-coloured products, and the soft Senonian rocks in Palestine do not give rise to red soil, in contrast to the harder Cenomanian rocks.

In view of these conflicting suggestions it was thought worth while to examine systematically some rendzina and red-brown soils and the underlying limestone rocks and in this paper is made a study of the chemistry of (i) rendzina and red-brown soils in order to ascertain the difference in exchange status, chemical composition, etc., and (ii) the insoluble silicate residue of the underlying limestone rocks to determine any relationship between the initial state of the parent materials of these two soil groups.

Experimental

Materials

Three rendzina profiles on chalk (Bu 8, Bu 20 and Bu 27), three red-brown soil profiles on limestone (So 104, So 140 and Dn 1), and two soil profiles, intermediate between red-brown and rendzina, overlying limestone (Y 172 and A 47), were collected from various limestone districts of England and Wales and one terra rossa profile on limestone (SF 2) was obtained from Southern France. The morphological description, topographic location, etc. of these profiles are given in Appendix I. They were selected from several other limestone soil profiles for their sedimentary origin as was evident from heavy mineral data for the soils and the insoluble residue of the underlying limestone sediment.⁸

Methods of determination

The soil and the insoluble residue of the underlying limestone were analysed chemically, the latter being determined by the method of Perrin.⁹ The soil and the limestone residue were fused with Na_2CO_3 as described by Piper¹⁰ and the following determinations were carried out on the melt : *silica* gravimetrically by the classical method ;¹⁰ *iron* colorimetrically by measuring the absorption of the Fe thioglycollate complex ;¹¹ and *aluminium* spectrophotometrically with ferron.¹²

Calcium and *magnesium* were determined by Versenate titration,¹³ in the case of the total soil on a HCl solution of the Na_2CO_3 melt, and for the limestone on an ammonium acetate extract after removal of acetate ions and destruction of organic matter.¹³

Rendzina soils contain an appreciable amount of free carbonate and hence these soils, excepting Bu 8/1 which has relatively lower amounts of carbonate, have not been studied for the distribution of exchangeable cations. The red-brown soils have also varying amounts of free carbonate (the data on the carbonate content of the different limestone soils are given in Appendix II, Table II). Because of the difficulties presented by the free alkaline earth carbonates, the selected profiles were analysed for *exchangeable cations* as follows : A double leaching with N-ammonium acetate was carried out on the same sample. The amounts of alkaline earth cations present in the second leachate were deducted from those of the first leachate, it being assumed that equal amounts of alkaline earths are released from free carbonate in the two leachates. This assumption may however be invalidated if the particle size distribution of the carbonates varies widely. The data obtained were thought to account for the exchangeable cations. The following cations were determined : calcium by Versenate titration ;¹³

magnesium spectrographically with ferric chloride as internal standard; and potassium and sodium on the EEL photometer. The cation-exchange capacity was determined by treatment of the NH_4^+ -saturated material with 10% aqueous NaCl, the displaced NH_4^+ ions in the leachate being determined by distillation in the Markham apparatus.¹⁴

Results

Tables I and II show the average values for the silica and sesquioxide content of different genetic soil types and the underlying limestone rocks and the chemical composition of the individual soil profiles is shown in Appendix II, Table I. The values have been averaged in Tables I and II to show the net genetic differentiation, if any, as there is sometimes overlapping of one genetic type with another when the individual profiles are considered. The terra rossa soil (SF 2/1) shows very close similarity with the red-brown soils and hence the data for them have been averaged with those of the latter.

Table I

Average values of silica and sesquioxides in different genetic soil types

Genetic type	Average of soil samples	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	SiO ₂	SiO ₂	SiO ₂
		g./100 g. of ignited soil (on alkaline-earth-free basis)			molar ratios		
Rendzina	Bu 8/1, Bu 20/1, Bu 20/2, Bu 27/1, Bu 27/2	62.65	17.40	8.97	4.94	6.45	25.16
Intergrade soil*	A 47/1, A 47/2, Y 72/1	69.13	14.86	11.24	6.70	5.56	15.36
Red-brown soil and terra rossa	So 104/1, So 104/2, So 140/1, So 140/2, Dn 1/1, SF 2/1	65.58	16.80	13.84	4.53	6.15	20.31

* Intergrade between red-brown soil and rendzina

Table II

Average values of silica and sesquioxides in the non-calcic residues of limestones

Rock type	Average of the limestone samples	SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	SiO ₂	SiO ₂	SiO ₂
		g./100 g. of ignited material (on alkaline- earth-free basis)			molar ratios		
Chalk underlying rendzina	Bu 8/2, Bu 20/3, Bu 27/3	55.74	21.06	6.93	3.77	4.59	22.30
Limestone underlying intergrade soil*	Y 172/2, A 47/3	50.01	21.20	8.00	3.22	6.12	47.09
Limestone underlying red- brown soil and terra rossa	So 104/3, So 140/3, Dn 1/2, SF 2/3	50.97	18.22	15.60	3.61	5.05	16.96

* Intergrade between red-brown soil and rendzina

Table III shows the exchangeable cations extracted by double leaching with ammonium acetate, and Table IV the net amount of exchangeable cations. Table III further shows that there is little release of alkali ions in the second leaching, indicating the presence of very little or no free alkali salts in the soils. No deduction of the alkali ions released in the second leaching has been made from the first leaching. Table IV shows a discrepancy between the NH_4^+ ions adsorbed and total exchangeable bases in some samples (So 140/1 and So 140/2). The high figure for the total exchangeable bases may be accounted for by a higher solubility of free carbonate in the first leachate and/or the fixed NH_4^+ ions being partly non-exchangeable with sodium ions. The degree of base saturation has been tentatively taken to be 100% in such samples. The terra rossa soil (SF 2/1) has not been analysed for exchangeable cations.

The data on the chemical characteristics of the different soil types are presented in Appendix II, Table II. Clay mineral distribution in the different soils and the underlying limestone rocks are given in Appendix II, Table III, for ready reference.

Table III

Exchangeable cations extracted by double leaching with ammonium acetate
(results as mequiv./100 g. of oven-dried soil)

Sample no.	Genetic type	First leaching (d ₁)				Second leaching (d ₂)			
		Ca	Mg	Na	K	Ca	Mg	Na	K
Bu 8/1	Rendzina	96.44	2.15	0.52	0.26	23.83	0.17	0.005	0.01
Y 172/1	Intergrade	29.33	3.74	0.14	1.06	14.03	1.75	0.010	0.04
A 47/1	"	96.37	2.47	0.62	0.97	13.25	0.40	0.005	0.05
A 47/2	"	55.13	1.75	0.38	0.84	13.96	0.30	"	0.01
So 104/1	Red-brown soil	41.45	1.33	0.27	0.66	8.39	0.19	"	0.005
So 104/2	"	39.62	0.78	0.20	0.31	9.52	0.06	"	"
So 140/1	"	61.19	1.96	0.84	0.49	17.55	0.17	"	"
So 140/2	"	63.87	1.30	0.64	0.29	25.87	0.19	"	"
Dn 1/1	"	43.57	1.24	0.73	0.73	8.39	0.22	"	0.03

Table IV

Exchangeable cations in different limestone soils (genetic types as in Table III)
(results expressed on oven-dried soil)

Sample no.	Ca	Mg	Na	K	NH ₄ ⁺	Total ex- change- able bases	Satura- tion %	Ca	Mg	Na	K	Carbon- ate as CaCO ₃ %
	(d ₁ -d ₂)*	(d ₁ -d ₂)*	(d ₁)	(d ₁)	ad- sorbed			% of total exchangeable bases	% of total exchangeable bases			
Bu 8/1	72.62	1.92	0.52	0.26	74.03	75.38	100	96.37	2.63	0.69	0.34	12.00
Y 172/1	15.30	1.99	0.14	1.06	19.74	18.49	93.69	82.74	10.76	0.76	5.73	26.87
A 47/1	50.12	2.07	0.62	0.97	58.4	29.78	100	93.89	3.46	1.04	1.62	2.70
A 47/2	41.17	1.45	0.38	0.84	46.88	43.84	93.52	93.91	3.31	0.87	1.92	4.80
So 104/1	33.06	1.14	0.27	0.66	46.06	35.13	76.27	94.12	3.25	0.77	1.88	0.44
So 104/2	30.10	0.72	0.20	0.31	46.88	31.33	66.83	96.08	2.30	0.64	0.99	0.27
So 140/1	43.64	1.79	0.84	0.49	43.59	46.76	100	93.35	3.83	1.80	1.05	4.68
So 140/2	38.00	1.11	0.64	0.29	32.49	40.04	100	94.92	2.77	1.00	0.72	13.11
Dn 1/1	35.18	1.02	0.35	0.73	43.59	37.28	85.52	94.35	2.74	0.94	1.96	2.73

* d₁ and d₂ as in Table III

Discussion

The data for the chemical composition of the different limestone soils (Table I) show that the genetic types can scarcely be differentiated by SiO₂, Al₂O₃, SiO₂/sesquioxide and SiO₂/Al₂O₃ ratios. The only distinctive features are the iron oxide content and SiO₂/Fe₂O₃ ratio. The composition of the non-calcic residues of the limestone rocks underlying the different genetic soil types (Table II) shows an exactly similar trend in the distribution of iron oxide as in the soils. This may be schematically shown as follows:

Rendzina	Increase → in Fe ₂ O ₃	Intergrade soil	Increase → in Fe ₂ O ₃	Red-brown soil
Chalk residue underlying rendzina	Increase → in Fe ₂ O ₃	Limestone residue underlying intergrade soil	Increase → in Fe ₂ O ₃	Limestone residue under- lying red-brown soil

This indicates that red-brown soil formation is connected with relatively more ferruginous residue of the hard limestones, whereas rendzina formation originates in a system with a lower amount of iron oxide.

The SiO₂/Fe₂O₃ ratio shows a higher value for rendzina and the corresponding underlying chalk residue, implying that the red-brown soils and the underlying limestone residues are more ferruginous and less siliceous than the other group of soils and rocks. An inspection of the SiO₂/Fe₂O₃ ratios further shows that this ratio is usually higher in the soil than in the rock, the difference being more marked in red-brown soil than in rendzinas. This possibly indicates a relatively greater degree of weathering and removal of iron oxide (with respect to silica), the process being more intensified with red-brown soil formation.

These findings on the chemical composition of the soil and rock (Tables I and II) therefore indicate that the difference between rendzina and red-brown soil is due to the difference in the composition of the initial non-calcic residues of the different limestone rocks, and possibly might not account for the genetic transformation of rendzinas into red-brown soils.

An inspection of Table IV shows that the red-brown soils are highly base-saturated (to the extent of 67–100%). The data for the carbonate contents (Appendix II, Table II) show that there is an appreciable difference in the state of decalcification between the rendzinas and red-brown soils, although the degrees of base-saturation are somewhat similar. A similar observation was also made by Stace¹⁵ working with some terra rossas and rendzinas of South Australia. From the foregoing it therefore appears that, while the rendzinas have a high base status with no decalcification, the red-brown soils are relatively more decalcified but maintain a high base status. This finding is not in agreement with that of Robinson¹ who observed that the red-brown soils are base-unsaturated. Robinson quoted low figures for exchangeable calcium and pH of a red-brown soil profile overlying carboniferous limestone at Bridgend, Glam., and discussed the degree of base-unsaturation and desilicification of red-brown soils. It may be mentioned that this low base status is not usual for red-brown soils or terra rossas as has been observed by the present author from a study on the exchange status of a number of red-brown soil and terra rossa profiles.⁸ Reifenberg,⁷ Nevros & Zvorykin⁴ and others have shown a base-saturated exchange complex of terra rossa soils. It seems therefore evident that in the formation of red-brown soils, the lowering of base status may be operative in only a few cases.

From an overall consideration of the findings it is concluded that Robinson's hypothesis¹ as to the formation of red-brown soils from the degradation (i.e., decalcification followed by desilicification) of rendzinas, may not be tenable. On the contrary, it has been found that the red-brown soils show a fair degree of correlation with the non-calcic residue of the underlying limestone. In other words the red-brown soils have been formed by the gradual accumulation of the insoluble residue of the limestone after dissolution of the bulk of the alkaline earth bicarbonates by weathering, the site of reaction being fairly well base-saturated.

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Appendix I

Soil profile descriptions

Rendzina soil profiles

Profile No. Bu 8

Locality	Summit of Ivinghoe Beacon, Bucks.
Series	Icknield
Type	Loam
Genetic group	Rendzina
Topography	Undulating. Site 750 ft. above sea level
Vegetation	Grass. <i>Festuca ovina</i> with yarrow (<i>Achillea</i>) and plantain
Underlying rock	Chalk rock (Upper Chalk)
Drainage	Excessive
Horizon	Depth, in.
A	0-7 Very dark brown (7.5 YR 3/2) humose loam; just moist; soft crumb structure; friable to loose but bound by abundant fibrous roots; occasional lumps of hard chalk and small calcareous particles.
C	7-12 Broken white chalk; hard and feels rather sandy.

Rendzina soil profile

Profile No. Bu. 27

Locality	Pitstonegreen Farm, Pitstone, Bucks.
Series	Wantage
Type	Clay loam
Genetic group	Rendzina
Topography	Gently undulating. Elevation 430 ft.
Vegetation	Arable
Underlying rock	Lower Chalk (marl)
Drainage	Free
Horizon	Depth, in.
A	0-7 Grey brown (2.5 Y 5/2) clay loam; very few small flints; weak subangular blocky; friable; abundant organic matter and roots; just moist; earthworms; very calcareous.
A/C	7-15 Light brownish grey (2.5 Y 6/2) clay; very few small flints and many chalk fragments; weak to medium subangular blocky; friable; some mechanical organic matter and roots; just moist.
C	15+ Broken chalk with soil from horizons above filling fissures. Depth of sampling: 1-4 in. (Bu 27/1); 9-12 in. (Bu 27/2); 16+ in. (Bu 27/3).

Profile No. So 140

Locality	Quarry on Creech hill, Som.
Series	Sherborne
Type	Clay loam
Genetic group	Red-brown soil
Topography	Level plateau
Vegetation	Old pasture, fine-leaved fescues probably dominant with cocksfoot, etc.
Underlying rock	Coarse-grained Jurassic limestone (Oolitic)
Drainage	Free
Horizon	Depth, in.
A	0-5 Reddish brown (5 YR 4/3) clay loam; soft granular structure; moist; some angular fragments of light yellowish-brown limestone; friable and mellow; moderate intimate organic matter; worms active; abundant grass roots slightly calcareous.

Profile No. Bu 20

Ninn Wood, Monk's Risboro', Bucks	
Icknield	
Loam	
Rendzina	
Undulating. 600 ft. O.D. Moderate slope, highly dissected terraces	
Beech; <i>Sanicula</i> dominant; moss	
Middle chalk with few flints	
Free to excessive	
Horizon	Depth, in.
A	0-4 Dark greyish brown (10 YR 3/1-2) loam with occasional small flints and chalk fragments; just moist; soft granular structure; very friable and porous; high intimate organic matter content (mull); litter space, no 'F' and 'H' layers.
A/C	4-13 Clear change to brownish grey (10 YR 5/3) silty clay loam with some dark grey-brown patches full of brown-stained chalk fragments; almost air-dry; fine material has a crumb structure; compact in place but very friable when removed; full of fissures; some intimate humus mainly in root tracks; roots much less frequent; extremely calcareous.
C	13+ Merging into broken white chalk with occasional brown staining passing into harder and more massive chalk below 18 in. (Sample Bu 20/3 from below 18 in.)

Red-brown soil profiles

Profile No. So 104

North Brottens, Doufing, Som.	
Sherborne	
Clay	
Red-brown soil	
Flat plateau feature	
Grassland, some clover, high herb content	
Fuller's Earth rock, Jurassic limestone	
Free	
Horizon	Depth, in.
A	1-5 Dark brown clay (10 YR 4/3-4); occasional small limestone fragments; strongly developed medium subangular blocky to granular structure; porous and finely fissured; friable; moderate intimate organic matter; turfy; fibrous roots; just moist.
(B)	5-14 Strong brown (7.5 YR 5/5) clay, with some duller coatings on structural elements; occasional angular limestone fragments; strongly developed medium subangular blocky structure; finely fissured; friable; low intimate organic matter; frequent fibrous roots; just moist.
C	14+ Broken limestone fragments.

Profile No. Dn 1

Near old telegraph house, Lllystacn, Denbighshire, North Wales.	
Gower	
Loam	
Red-brown soil	
Undulating. Elevation 600 ft.	
Permanent grassland	
Carboniferous limestone	
Free to excessive	
Horizon	Depth, in.
A	0-5 Dark reddish brown (5 YR 3/3) loam; frequent limestone fragments; crumb structure; porous; high to moderate intimate organic matter content; very abundant roots forming close network; moist.
C	5+ Hard crystalline limestone.

(continued)

Horizon	Depth, in.	
(B)	5-10	Yellowish-red clay containing abundant red-brown stained angular limestone fragments; moist; granular; friable.
C	10+	Broken coarse-textured oolitic limestone, mainly horizontally bedded, but irregular and rubbly. (limestone sampled at 24-30 in.).

Soil profiles intergrade between rendzina and red-brown soils

Profile No. Y 172

Locality	Wetherby, Yorks.
Series	Aberford
Type	Loam
Genetic group	Rendzina to red-brown soil
Topography	Undulating. Elevation 125 ft.
Vegetation	Old pasture
Underlying rock	Magnesian limestone (Permian)
Drainage	Excessive

Horizon	Depth, in.	
A	0-7½	Fine sandy light loam; dark grey (10 YR 3/1.5) in colour; subangular blocky to crumb structure; some limestone fragments; abundant roots and moderate intimate organic matter content.
C	7½+	Limestone rock.

Profile No. A 47

Locality	Bwrdd Arthur (Dinas Sylwy), Anglesey, North Wales
Series	Gower
Type	Loam
Genetic group	Rendzina to red-brown soil
Topography	Undulating. Elevation 450 ft.
Vegetation	Grassland
Underlying rock	Carboniferous limestone
Drainage	Excessive

Horizon	Depth, in.	
A	0-3	Dark grey brown (10 YR 3/2, dry) loam; occasional limestone fragments; fine crumb structure; porous; friable; much intimate organic matter; roots abundant; slightly moist.
(B)	3-8	Brown (10 YR 3/2, dry) loam; numerous limestone fragments; porous; friable; moderate intimate organic matter; slightly moist.
C	8+	Limestone rock.

Terra rossa soil profile

Profile No. SF 2

Locality	Near Vallone, Provence, S. France
Type	Clay
Genetic group	Terra rossa
Topography	Undulating
Vegetation	Grass, pines, etc.
Underlying rock	Muschelkalk rock, Triassic limestone
Drainage	Free

Horizon	Depth, in.	
A	0-½	Leaf litter. No differentiation into 'F' and 'H' layers.
A	½-4	Red clay (2.5 YR 3/4-dry) with a greyish tint; fair amount of intimate organic matter; strongly developed polyhedral structure; few carbonate fragments; friable when dry, sticky when moist.
(B)	4-8	Red clay (2.5 YR 3/6-dry); occasional carbonate fragments; decomposed plant roots; prismatic structure development (season of sampling, desiccating summer); sticky when moist.
C	8+	Limestone rock.

Appendix II

Chemical properties and clay mineral distribution

Table I

Chemical composition of the different soil types and the underlying limestone rocks

Sample no.	Genetic type	g./100. g. of ignited material							Molar ratios		
		SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	CaO	MgO	SiO ₂ R ₂ O ₃	SiO ₂ Al ₂ O ₃	SiO ₂ Fe ₂ O ₃		
Bu 8/1	Rendzina	38.40	17.15	9.12	19.04	4.68	2.85	3.82	11.25		
Bu 8/2*	"	1.70	0.61	0.30	91.43	5.76	3.41	4.41	14.95		
Bu 20/1	"	15.76	4.46	1.76	70.84	4.38	4.77	5.98	23.60		
Bu 20/3*	"	3.02	1.38	0.37	91.80	8.54	3.28	3.75	26.63		
Bu 27/1	"	31.35	6.28	1.67	54.08	1.38	7.37	8.57	52.30		
Bu 27/2	"	27.52	6.34	5.44	56.45	1.96	4.77	7.41	13.41		
Bu 27/3*	"	12.75	3.84	1.35	70.05	4.97	4.63	5.61	25.35		
Y 172/1	Intergrade**	60.02	6.46	4.06	16.37	15.61	11.29	4.83	8.65		
Y 172/2*	"	1.16	0.40	0.49	55.49	36.78	3.10	8.78	43.56		
A 47/1	"	54.04	17.84	6.26	8.68	1.73	4.21	5.15	22.89		
A 47/2	"	54.53	13.92	9.94	6.38	1.28	4.59	6.70	14.55		
A 47/3*	"	5.16	2.54	0.28	88.65	1.87	3.33	3.46	50.62		
So 104/1	Red-brown soil	58.81	15.61	17.87	2.97	1.78	3.70	6.41	8.75		
So 104/2	"	58.50	11.04	13.86	2.60	1.33	5.01	9.05	11.23		
So 104/3*	"	4.89	1.22	1.88	88.34	2.57	4.25	6.82	11.28		
So 140/1	"	59.74	15.44	9.87	9.56	2.57	4.68	6.60	16.33		
So 140/2	"	51.66	12.85	20.11	13.22	2.24	3.41	6.85	6.80		
So 104/3*	"	1.91	1.19	1.65	95.50	2.50	1.45	2.67	3.20		
Dn 1/1	"	66.00	15.12	6.72	5.64	2.57	6.22	7.99	28.10		
Dn 1/2*	"	0.82	0.24	0.58	96.07	0.53	4.96	5.71	38.05		
SF 2/1	Terra rossa	62.33	21.20	6.72	5.98	4.79	4.16	5.00	24.74		
SF 2/3*	"	1.56	0.53	0.27	45.93	48.25	3.77	5.00	15.29		

* Underlying limestone

** Intergrade between red-brown soil and rendzina

Table II

Chemical characteristics of the different limestone soils
(results on oven-dry basis)

Sample no.	Genetic type	Organic C, %	Organic N, %	Loss on ignition %	Carbonate as CaCO ₃ %	pH (in CaCl ₂)
Bu 8/1	Rendzina	13.06	1.15	26.82	12.00	7.3
Bu 20/1	"	7.81	0.90	17.10	69.80	7.6
Bu 27/1	"	2.15	0.24	7.80	59.00	7.6
Bu 27/2	"	0.80	0.11	2.86	66.72	7.8
So 104/1	Red-brown soil	6.46	0.68	19.10	0.44	6.2
So 104/2	"	3.97	0.42	14.48	0.27	6.3
So 140/1	"	8.35	1.16	22.10	4.68	7.2
So 140/2	"	5.18	0.66	14.98	13.11	7.3
Dn 1/1	"	11.25	0.95	24.96	2.73	6.9
Y 172/1	Intergrade*	5.01	0.49	10.81	26.87	6.9
A 47/1	"	11.89	1.69	29.78	2.70	6.6
A 47/2	"	11.53	1.23	26.53	4.80	7.1
SF 2/1	Terra rossa	1.50	0.30	4.87	Trace	5.6

* Intergrade between red-brown soil and rendzina

Table III

Clay mineral distribution in different soils and underlying limestone rocks

Sample no.	Genetic type	Mica	Kaolin	Montmorillonite	Vermiculite	Goethite
Bu 8/1	Rendzina	Little	Moderate	Moderate	---	Trace
Bu 8/2	Upper chalk	Moderate	Very little	"	---	---
Bu 20/1	Rendzina	"	"	Dominant	---	Trace
Bu 20/3	Middle chalk	"	---	"	---	---
Bu 27/1	Rendzina	"	Very little	"	---	---
Bu 27/3	Lower chalk	"	Little	"	Very little	---
Y 172/1	Intergrade soil*	"	Moderate	---	Little	Trace
Y 172/2	Permian limestone	Dominant	Very little	---	---	Very little
A 47/1	Intergrade soil*	Moderate	Moderate	---	Moderate	Trace
A 47/3	Carboniferous limestone	Dominant	"	---	---	---
So 104/2	Red-brown soil	Moderate	Little	---	Dominant	Trace
So 104/3	Jurassic limestone	Dominant	"	---	Very little	"
So 140/1	Red-brown soil	Little	"	---	Moderate	Very little
So 140/3	Jurassic limestone	Dominant	"	---	Little	Trace
Dn 1/1	Red-brown soil	Moderate	"	---	Moderate	"
Dn 1/2	Carboniferous limestone	Dominant	Very little	---	---	Very little
SF 2/1	Terra rossa	Moderate	Moderate	---	Moderate	Trace
SF 2/3	Triassic limestone	Dominant	Little	Little	Very little	"

(from X-ray data)

* Intergrade between red-brown soil and rendzina

Trace = 0-4%
Very little = 5-10%
Little = 11-25%
Moderate = 26-50%
Dominant = 50%

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ABSTRACTS

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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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JOURNAL OF THE SCIENCE OF FOOD AND AGRICULTURE ABSTRACTS

AUGUST, 1960

I.—AGRICULTURE AND HORTICULTURE

General: Soils and Fertilisers

Characteristics of soil formation on early pleistocene boulder clays in Schleswig-Holstein. H. E. Stremme and H. Bach (*Z. PflErnähr. Düng.*, 1960, **88**, 148—155).—In these soils the clay content is higher in the B than in the A or C horizons and higher in the upper than in the lower B horizon. T-values are in agreement with this. Arable soils have a higher base saturation and pH than woodland.

M. LONG.

Automatic recording of soil moisture and other ecological factors. R. D. Frazier and A. R. Bertrand (*Agron. J.*, 1960, **52**, 53—54).—The equipment is described.

A. H. CORNFIELD.

Rates of water entry into a chernozem soil as affected by age of perennial grass sod. A. P. Mazurak, W. Kriz and R. E. Ramig (*Agron. J.*, 1960, **52**, 35—37).—Rate of water entry into a 2-year-old grass sod (silt loam chernozem) was not much different from that into soil which had been in grain-fallow for 5 years. Water entry into a 4-year-old sod was significantly greater than that into grain-fallow soil. Water entry increased somewhat with age of grass sod, but there was little difference between an 8- and a 20-year-old sod. Application of N to the grass did not affect water entry. Bulk density of the surface 3 in. decreased with increasing age of grass stand. Air-filled pore space at 60 cm. tension or at field capacity was not related to age of grass stand.

A. H. CORNFIELD.

Movement of water in soils using tritium-labelled water. H. W. Scharpenseel and H. Gewehr (*Z. PflErnähr. Düng.*, 1960, **88**, 35—49).—³H studies show that water movement reaches 1 m. in depth within one day. Where an impermeable layer exists, the activity of the layers beneath it reaches a peak and falls off, as the water moves more rapidly through the more permeable layer. Horizontal spreading lags behind vertical movement, but in the upper 30-cm. layers spreading to at least 30 cm. from point of application occurs, although the surrounding area is kept at the same water status. Worm holes have a marked effect on soil hydrology.

M. LONG.

Exchange capacity of soils and various soil constituents in Finland. R. Heinonen (*Z. PflErnähr. Düng.*, 1960, **88**, 49—59).—The base-exchange capacity (B.E.C.) of fractions $>1\mu$ is very small; that of clay particles $<1\mu$ is ≈ 40 mequiv./100 g. in glacial soils in which the humus content has no influence on the total capacity. In light soils of low humus content the B.E.C. averages 300 mequiv./100 g. declining with increasing humus content to approx 200 mequiv in well-humified low-moor peats. In general the B.E.C. of soil org. matter decreases with rise in clay content of the soil.

M. LONG.

Effect of colloidal silica in peptising iron oxide with reference to red brown soil formation on limestone. D. H. Khan (*J. Sci. Ed Agric.*, 1960, **11**, 133—136).—The peptisation of cold pptd. and ignited forms of Fe_2O_3 by alkaline silica sol occurs in presence of Na^+ but not in alkaline earth media. Ca^{2+} precipitates silica from the dispersed phase. The fate of silica liberated by weathering of limestone is governed by the Ca^{2+} and Mg^{2+} saturation of the system. Reifenburg's hypothesis on the formation of terra rossa on limestone is untenable. (13 references.)

E. M. J.

Nitrate content of soils and nitrogen content of oat plants as affected by rates of liming. C. Ogata and A. C. Caldwell (*Agron. J.*, 1960, **52**, 65—68).—Liming an imperfectly-drained silt loam increased NO_3^- accumulation in the fallow soil. Soil- NO_3^- increased from May to Sept. and decreased thereafter, probably due to heavy rainfall. By Sept. fallow soil had accumulated 600—1200 lb. of NO_3^- per acre in the 0—24 in. layer. Use of 16 tons per acre of $CaCO_3$ was no more effective than was 4 tons per acre in increasing soil- NO_3^- content. When the soil was cropped with oats, soil- NO_3^- content usually did not exceed a few lb. per acre and liming had little effect on these values. Liming had no consistent effect on the N content of oat plants.

A. H. CORNFIELD.

Availability of soil inorganic phosphate as measured by plant uptake and adsorption by anion-exchange resin. P. G. Orth (*Dissert. Abstr.*, 1959, **20**, 1945).—Sudan grass took up about the same

amount of P from two different soils in greenhouse experiments. Addition of P fertiliser increased P uptake and crop yield more on the soil with pH 6.4 than on that with pH 5.6. An anion-exchange resin equilibrated with the soils adsorbed more P than was available to the crop, both before and after fertilising and cropping.

M. D. ANDERSON.

Potassium economy and mineral status of Göttingen E-fields. F. Scheffer, E. Welte and H. Graf v. Riechenbach (*Z. PflErnähr. Düng.*, 1960, **88**, 115—128).—The total K contents of these soils lie between 1.78 and 1.94%, 41.8% of this occurring mainly as feldspar in the fine sand fraction, 24.4% as mica in the silt fraction and 28.2% in the clay fraction. The K in the first-named two fractions is non-available; that in the clay occurs as inter-lattice K (I) in illite clay minerals. About 1.3% of the total K is exchangeable (II). In a plot receiving no K I falls more rapidly than II. After exhaustion of I the progressive supply from non-available K sources may not be sufficient for crop requirements.

M. LONG.

Paper chromatographic investigations of humic matter, humic acid and model compounds. W. Scharpenseel (*Z. PflErnähr. Düng.*, 1960, **88**, 97—115).—The more rapidly moving humic acid decomposition products have R_f values almost identical with those of products resulting from the alkaline oxidation of quinol and pyrogallol. Decomposition products, arising from alkali fusion, nitrobenzene oxidation, acid hydrolysis, n-butanol extraction and neutralisation by quaternary ammonium bases of humic acid prep. include pyrocatechol, pyrogallol, protocatechuic acid and isophthalic acid. Exhaustive extraction of humic acid with n-butanol yields pyrrole-2-carboxylic acid and pyrogallol-5-carboxylic acid. Grey and brown humus have fundamentally similar chemical components.

M. LONG.

Soil aggregation and organic-matter decomposition. G. Chesters (*Dissert. Abstr.*, 1959, **20**, 1961).—Water-stable soil aggregates were increased by addition of org. substances that decay rapidly, but not by those that decay slowly. Addition of N with the rapidly decomposing substances decreased aggregation. The polysaccharide content of soil was closely correlated with degree of aggregation. The ratio of roots to tops in crops of carrots and rape was used to assess effects of aggregation on growth; there was an almost entire absence of secondary root growth in puddled soils. Org. acids and their deriv. in soil were characterised by differential thermal analysis, employing free access of air. The thermal patterns of bentonite, haemoglobin and glycine, and of bentonite complexes with haemoglobin and glycine, suggested that although the substances are themselves hydrophilic, the complexes are hydrophobic. Cellulose and wood sawdust gave similar thermograms, but isolated spruce lignin gave a different pattern.

M. D. ANDERSON.

Activity of enzymes and respiration intensity in soils. A. Sh. Galstyan (*Dokl. Akad. Nauk SSSR*, 1959, **127**, 1099—1102).—A simple apparatus for measuring soil respiration is devised and the enzyme activity in cultivated black chernozem, dark chestnut, medium chestnut, light chestnut, brown, and light brown soils is examined. The chernozem showed a high level of invertase, amylase, β -glucosidase and urease, and comparatively low levels of respiration intensity, peroxidase, polyphenoloxidase and catalase. In the brown soils this order of activity was reversed. In general, in the dark soils the hydrolytic were more intense than the oxidation-reduction processes and in the lighter coloured soils vice versa.

A. L. GROCHOWSKI.

Relative sensitivity of nitrifying bacteria to hydrogen ions in soils and in solutions. D. F. Weber (*Dissert. Abstr.*, 1959, **20**, 1975).—The nitrifying organisms of acid soils continued to produce nitrates even after the pH had fallen to 4.0. N as $(NH_4)_2SO_4$ tended to be toxic to nitrification in soil, but was readily transformed to nitrate when present in a buffered nutrient solution, perfused through columns of soil; final pH values were 3.9 to 4.1. Nitrification did not occur in agitated liquid media at pH below 6.0.

M. D. ANDERSON.

Denitrifying flora of soil. C. L. Valera (*Dissert. Abstr.*, 1959, **20**, 1929—1930).—The no. of denitrifying bacteria in a sandy loam soil were correlated with pH. The optimum pH was 6—8 for five spp. of denitrifying bacteria; their range of tolerance varied, but denitrification by four spp. was inhibited at pH 4.0 and 9.8. When

NH₄Cl, NH₄NO₃ or KNO₃ was added to the soil at neutral pH, nitrification rapidly brought the pH to about 4.5, owing to the low buffering capacity of the soil. The denitrifiers could be classified in four groups, (i) those denitrifying with NO₃⁻ as sole source of N, (ii) those requiring NH₄⁺, (iii) those requiring amino-acids, and (iv) those requiring high concn. of amino-acids. Only 3% of denitrifiers were aerobic spore-formers. M. D. ANDERSON.

Role of calcium and acetate in nitrogen fixation by *Azotobacter vinelandii*. J. A. Bush (*Dissert. Abstr.*, 1959, 20, 1960—1961).—*Azotobacter vinelandii*, strains O and OP, require some Ca for optimum growth with combined N, and a markedly higher Ca concn. for optimum N fixation. Acetate can replace the Ca needed for N fixation, but does not stimulate growth with fixed N. Ca does not stimulate oxidation of sucrose by Ca-deficient cells of strain OP. Trace quantities of acetate initiate N fixation of strain OP in the absence of Ca, and fixation continues after exhaustion of the acetate, with sucrose as substrate. ¹⁴C from labelled acetate occurs principally in the protein fraction of cells after incubation for 6 hr. A. M. SPRATT

Availability of bacterial cell substances as nutrient material for other organisms. H. C. Mussman (*Dissert. Abstr.*, 1959, 20, 1965—1966).—Ruptured bacterial cells used as a nutrient for other bacteria and a sp. of protozoa increased growth in each case. With germinated wheat the cells did not affect blade growth, while root lengths varied inversely with substrate concn. A. M. SPRATT.

Effect of nitrate of soda on the physical condition of the soil: in warm-climate arid agriculture. H. Nicol (*Chil. Nitrate agric. Serv. Inform.*, 1959, Nov., 7 pp.).—The idea that NaNO₃ is injurious to the physical condition of alkaline or saline soils in tropical or subtropical regions and the contrary view that NaNO₃ is to be preferred to NH₄⁺ fertilisers which lose N on such soils, are discussed. NaNO₃ in all soils has a high value in maintaining an ionic balance favourable to growth of crops. E. M. J.

Keeping quality of fertilisers. II. Caking and hygroscopicity of calcium ammonium nitrate. S. Varma and K. R. Chakravorty (*J. sci. industr. Res.*, 1959, 18B, 486—488).—Comparative data on the hygroscopicity of diluted and pure NH₄NO₃ are given. CaNH₄ nitrate (I) and nitro-kaolin are about twice as hygroscopic as NH₄NO₃ on the basis of NH₄NO₃ content. A coating of 6% chalk on I granules reduces the caking tendency. I. JONES.

Fertiliser nitrogen and its loss to atmosphere. R. A. Schwartzbeck (*Dissert. Abstr.*, 1959, 20, 1947).—Applications of 0—1080 lb. of N per acre as NH₄NO₃ did not affect yields of maize grain. Leaf-N was highest 2—3 weeks after application. Supplements of K and P increased the N content of the grain. Analyses of the gases evolved from soils treated with N fertilisers labelled with ¹⁵N showed only trace losses of N₂O and/or N₂, except with water-saturated soils treated with NH₄NO₃ or HNO₃; practically all the evolved N₂ came from the NO₃ group. More N₂O was evolved from one water-saturated soil than could have come from fertiliser N. More N₂O than N₂ was usually evolved, except when N was applied as HNO₃. High loss of N from HNO₃ was associated with high content of org. matter in the soil. M. D. ANDERSON.

Gaseous loss of ammonia from nitrogen fertilisers applied to soils. C. B. Kresge and D. P. Satchell (*Agron. J.*, 1960, 52, 104—107).—When applied at rates >100 lb. of N per acre there were fair losses of N as NH₃ from application of urea and cyanamide to the surface of bare soil. When top-dressed on Coastal Bermuda grass, losses of NH₃ from urea were much less than those from bare soil. Losses of NH₃ from urea were reduced by watering it into the soil or by preventing evaporation of soil moisture. Mixing urea with NH₄NO₃ (I) in conc. solutions before application reduced the amount of NH₃ lost. (NH₄)₂SO₄ and I applied even at 300 lb. per acre showed no loss of NH₃. A. H. CORNFIELD.

Residual response to urea fertilisation. E. G. Cuthbertson (*J. Aust. Inst. agric. Sci.*, 1959, 25, 295—296).—Urea applied to a wheat crop in a drought year produced a considerable increase in yield of oats in the following season. A. G. POLLARD.

Useful correlations between results of fertiliser trials and other field data. C. B. Wells (*J. Aust. Inst. agric. Sci.*, 1959, 25, 273—281).—Observations in S. Australia on soils deficient in and those adequately supplied with K, Mn, Cu, Zn and Mo show a distinct geographical distribution differing for each element. Such distributions are also related to soil type and to underlying geological formations. Use of this data in extending the findings of local field trials to cover larger areas is discussed. A. G. POLLARD.

Straw and composts. I. Characterising straw, composts and bulky organic manures by optical extinction of alkaline extracts and cation-exchange capacity measurements. A. H. Cornfield (*J. Sci. Fd Agric.*, 1960, 11, 125—128).—Both methods are suitable for measuring the

extent of humification of rotted materials. Of extracts made with 0.05N-NaOH (containing 0.5% of "Calgon") on straw, composts prepared in various ways, farmyard manure and peat, straw extracts had the lowest and peat extracts the highest extinction value and cation-exchange capacity. The high correlation between the two methods (with materials of widely different sources) indicates that the differences are due to no. rather than nature of chemical groups present, exhibiting exchange properties. (10 references.) E. M. J.

Application of thermogravimetry to the analysis of carbonates occurring in soils. I. Analysis of pure carbonates and naturally occurring limestones. J. R. Wright, I. Hoffman and M. Schnitzer. **II. Analysis of carbonates in soils.** I. Hoffman, M. Schnitzer and J. R. Wright (*J. Sci. Fd Agric.*, 1960, 11, 163—167, 167—172).—I. Analyses of carbonates, pure and in mixture and of naturally occurring limestones, in an atm. of CO₂ were studied. Thermogravimetry in an atm. of CO₂ permits accurate determinations of total carbonates and the partition into calcite and dolomite in one operation. The results were compared with those by chemical and X-ray diffraction methods. (11 references.)

II. Values for total carbonates were in good agreement with those obtained by chemical methods. Partitioning of total carbonate into dolomite and calcite appeared possible from examination of some curves. For most of the soils where this was possible, results for dolomite were higher and those for calcite lower, than values obtained by a chemical method. The interaction during pyrolysis, between various inorg. salts and quartz with calcite was studied. Difficulties in partitioning were caused by interaction between Na or K and SiO₂ with calcite, and caution is needed in interpreting the results. E. M. J.

Complexometric determination of calcium and magnesium. E. Rauterberg, H. Ossenberg-Neuhaus and A. Wiegboldt (*Z. PflErnähr. Düng.*, 1960, 88, 14—17).—Fe³⁺, Al³⁺ and PO₄³⁻ are pptd. before the titrations by addition of FeCl₃ followed by a gradual neutralisation of the solution up to the turbidity point. An acetate buffer at pH 4.6 is added and the interfering ions filtered off. A murexide-Naphthol Green indicator is used for the Ca titration and an Eriochrome Black-methyl orange indicator for the Mg. M. LONG.

Determination of potassium in fertilisers by flame photometry. N. Mevel, J. Angot and L. Vanoverberghe (*Chim. anal.*, 1960, 42, 15—23).—Disadvantages of gravimetric methods for K are discussed. Flame photometry of K in fertilisers can be effected if a cool flame (butane-air) is used (suppressing interference from foreign cations) and the reading is taken for the 766.5/770 mμ doublet with an interference filter. Interference of sulphate ion is overcome by having sulphate present in the standard. Foreign cations are not present in sufficient quantities to interfere. E. J. H. BIRCH.

Examination of agricultural and horticultural products by the formic acid technique to determine cellulose, starch and protein. E. Lehmann and H. Birsgal (*Z. PflErnähr. Düng.*, 1960, 88, 1—13).—The material is treated with hot to remove fats, solids being separated by centrifuge. These are then treated with formic acid 16—20 hr. for material of high and 40—50 hr. for that of low starch content. Starch, cellulose, protein and simple saccharides and amino-compounds are thus formylated. Subsequently the cellulose derivative is pptd. and removed in the centrifuge, the formylated starch is pptd. by acetic acid and benzene and the protein derivative by ether, benzene and light petroleum, leaving the simpler compounds in solution. Deformylation is carried out on the cellulose with 1.6% NaOH or "Cuoxam," on the starch with 5% NaOH for 6 hr. followed by acidification with acetic acid and pptn. with methanol, and on the protein by treatment with 2% aq. pyridine, acidification with acetic acid and pptn. with acetone. Results for various types of produce are given. M. LONG.

Composition for temporarily sterilising soil. Stauffer Chemical Co. (B.P. 789,690, 22.7.55. U.S., 28.7.54 and 22.6.55. Amended 2.2.59).—An aq. stable concentrate (which after dilution with water is used for the temporary sterilisation of soil) contains 20—40 wt.-% of a compound NHR-CS₂M (R is Et or Me; M is alkali metal, alkaline-earth metal or NH₄) and 0.1—1 wt.-% of an amine. Thus, an aq. solution containing 40% of Na methylthiocarbamate is only decomposed to the extent of 4.2% (after 1 week at 60°) as compared with 68.5% for a 2.5% aq. solution, and when applied to soil at a concn. of 100 p.p.m. killed wireworm larvae. When seeds were planted 7 days after the application they grew without wireworm damage. F. R. BASFORD.

Plant Physiology, Nutrition and Biochemistry

Relation of sugar content to frost-hardiness in plants. A. Sakai (*Nature, Lond.*, 1960, 185, 698—699).—The extent to which frost-

hardness of the parenchyma cells of the cortex of the mulberry is increased by chilling is proportional to the increase in the concn. of sucrose in the cells. Frost-hardiness in mulberry twigs cut in April was increased by gradual desiccation at room temp., which caused decrease in water and starch contents, and increase in sugar and osmotic concn. In twigs frost-hardened by chilling and held at 15° for 20 days without desiccation to use up starch granules in the parenchyma cells, further chilling did not increase frost-hardiness or sugar content. Increase in sugar concn. may be a primary factor in frost-hardiness, on which chilling as such has no direct effect. Twigs of gardenia put in glucose solution for 24 hr. at 25° showed increased sugar concn. and frost hardiness. Other sugars, polyols and acetamide, but not inorg. salts, had the same effect.

M. D. ANDERSON.

Magnesium-supplying power of glazed porcelain pots. F. J. Roberts (*J. Aust. Inst. agric. Sci.*, 1959, **25**, 298—299).—Plants grown in glazed porcelain pots obtained significant amounts of Mg from the pots. Talc was used in the manufacture of the pots.

A. G. POLLARD.

Calcium-lithium competition in absorption by plant roots. E. Epstein (*Nature, Lond.*, 1960, **185**, 705—706).—The absorption of Li by excised barley roots was competitively inhibited by Ca. Ca accelerated the absorption of Rb like that of K. Li ions resemble the ions of the alkaline earths more than they resemble other alkali ions in some of the properties that determine chemical affinities, including affinity for the carrier sites involved in the active transport of ions across cell membranes.

M. D. ANDERSON.

Accumulation of caesium-137 by plants grown in simulated pond, wet meadow and irrigated field environments. R. C. Pendleton and R. L. Uhler (*Nature, Lond.*, 1960, **185**, 707—708).—Plants of *Polygonum persicaria* were grown outdoors in conditions simulating shallow ponds, wet meadows, and irrigated fields. The uptake of ¹³⁷Cs by the plants, measured 36 days after application to the surface of the soil or water, was in the ratio 450 : 30 : 1, respectively. In connexion with fall-out, this uptake by roots would be additional to absorption from aerial plant surfaces.

M. D. ANDERSON.

Factors influencing radio-strontium uptake by plants. E. M. Romney, W. L. Ehler, A. H. Lange and K. H. Larson (*Plant & Soil*, 1960, **12**, 41—48).—The uptake of ⁹⁰Sr from a loam varied considerably with plant species. In the mature plant top turnip and millet showed the highest and spinach the lowest concn. of Sr. In cereals the Sr concn. in the grain was about 20% of that in the forage, whilst potato tubers contained about 2% of that in the tops. In culture tests the uptake of Sr by barley and beans was reduced by decreasing temp., light intensity or light exposure period.

A. H. CORNFIELD.

Non-medical uses of antibiotics. II. Horticulture. R. Levin (*Chem. Prod.*, 1960, **23**, 157—160).—The use of cycloheximide, griseofulvin, nystatin, antimycin and helixin are particularly discussed and the mode of action and formulation are indicated. Induced sensitisation and microbial resistance are considered together with the persistence of these substances within the produce.

C. V.

Action of streptomycin on the growth of crops. H.-O. Leh (*Z. Pflernähr. Dting.*, 1960, **88**, 129—148).—Peas and common vetch in hydroponic and cucumber and maize in soil cultures are favourably affected by streptomycin in the media, but high concn. depress growth and may cause death. Chlorosis is caused in peas, necrosis in vetch and chlorosis and marked axillary shoot growth in cucumber. Soya-bean and sunflower are adversely affected even at low concn.; necrosis occurs in the former, chlorosis in the latter. Cucumber, sprayed with streptomycin, grow more side shoots and the no. and sex of flowers are affected. Mg-deficiency symptoms appear in summer aster repeatedly sprayed with streptomycin. This can be remedied by spraying with MgSO₄.

M. LONG.

Inhibition of rooting by maleic hydrazide. P. May (*J. Aust. Inst. agric. Sci.*, 1959, **25**, 304—306).—Placing sultana cuttings in a solution of maleic hydrazide (12 p.p.m.) for 24 hr. before potting restricted root development. Higher concn. (50 p.p.m.) prevented rooting entirely; with 3 p.p.m. no effects were apparent.

A. G. POLLARD.

Gibberellic acid and the chilling requirements of peach seeds. M. G. Mes (*Nature, Lond.*, 1959, **184**, 2034—2035).—Peach seeds normally require 2 months at low temp. before they germinate. Removal of the seed coats permits germination without chilling, but the seedlings show growth abnormalities. Attempts to induce germination of unchilled seeds, or normal growth of embryos excised from the seed coats by various concn. of gibberellic acid applied by soaking the seeds, application to the growing points, spraying, or in lanolin, were all unsuccessful. Unchilled seeds placed in agar-containing gibberellic acid showed some improvement in % germination but only dwarf seedlings developed.

M. D. ANDERSON.

Influence of gibberellic acid and photoperiod on the growth, flowering, nodulation and nitrogen assimilation of *Vicia villosa*. M. G. Mes (*Nature, Lond.*, 1959, **184**, 2035—2036).—The effects of gibberellic acid on *Vicia villosa* varied with the age of the plants. In plants grown in sand culture with a N-free nutrient, gibberellic acid in the first 3 weeks increased the length without increasing the dry wt. or N content; root growth and nodulation were poor. After 6 weeks, the increase in wt. was the same in treated and control plants, but the latter had higher dry wt. and % N. The dry wt. and total N content of well-established plants were increased by gibberellic acid when the photoperiod was short, but not when it was long. In still older plants, gibberellic acid had little or no effect except that height was increased.

M. D. ANDERSON.

Influence of hydrogen ion concentration and autoclaving on gibberellin. J. H. M. Henderson (*Nature, Lond.*, 1960, **185**, 628—629).—Gibberellin solutions on autoclaving lose 25—33% of their activity (as measured by the growth in length of the second internode of dwarf-1 mutant maize during the 2 weeks after application of gibberellin). The activity of solutions after autoclaving was greatest in those at pH 3.5 and 6.0, and least in those at pH 1.0, with those at pH 10.0 intermediate. The loss of activity at the normal pH of culture media is not enough to be of importance experimentally. Other growth factors would show approximately similar losses.

M. D. ANDERSON.

Crops and Cropping

Effects of elevated atmospheric pressures on growth of wheat plants. H. R. Gardner and H. M. Taylor (*Agron. J.*, 1960, **52**, 23—25).—Wheat plants grew normally under air pressure of 3 atm., but growth rate decreased with further increasing atm. pressure. At 7—9 atm. there was no further growth after 3—4 cm. and the plants were extremely chlorotic.

A. H. CORNFIELD.

Excessive soil moisture: effect on barley plants in various stages of development. N. N. Savitskaya (*Dokl. Akad. Nauk SSSR*, 1959, **128**, 850—852).—The effects of 100% moisture saturation of soil and of soil flooding at the four main stages of the growth of barley on the grain yields were tabulated. Flooding of soil in the early and intermediate stages of plant development considerably decreased the grain crop. Flooding at the initial pre-exposed growth stage reduced the yields only to 78% of the controls, at the next exposed formative stage to 35%, and at the third stage prior to the formation of tetrads in the anthers to 55%. The effects of 100% moisture saturation of the soil were less drastic. Flooding during the initial growth of the barley increased the bound and decreased the free water contents in the plants. These changes affected the functions of roots, plant respiration and oxidation processes. Flooding the soil in the final stage of growth and 100% moisture saturation at all stages had little effect on the free or bound water in the tissues. (12 references.)

A. L. GROCHOWSKI.

Effect of nitrogen on non-irrigated maize sown in pasture slits. J. Strang and P. Broue (*J. Aust. Inst. agric. Sci.*, 1959, **25**, 312—314).—Maize was sown in slits made in a perennial ryegrass pasture. Various split applications of (NH₄)₂SO₄ significantly increased grain yields, wt. of ears per 100 plants, wt. per ear, 1000-grain wt. and % N in grain. Similar trials in kikuyu grass (*Pennisetum clandestinum*) and paspalum pastures were unsuccessful due to competition of the grasses for N and water.

A. G. POLLARD.

Fixation of nitrogen by the rice plant. S. P. Chakraborty and S. P. Sen Gupta (*Nature, Lond.*, 1959, **184**, 2033—2034).—Rice seedlings 3 weeks old were grown in nutrient solution for 4 weeks, and their average N content was determined. Some were then transferred to N-free media. After a further 4 weeks, the average N content of these had increased by about 50%. Seedlings transferred to N-free media at 8 weeks showed an average increase of N content of 6—7% in the next 4 weeks. *Azotobacter* were absent from the nutrient solutions after the periods of N starvation.

M. D. ANDERSON.

Nutrition of rice plant (*Oryza sativa*, L.). VII. Influence of increasing levels of phosphate and potash on growth, yield and nutrient uptake by rice plant. A. Tanaka, S. Patnaik and C. T. Abichandani (*Proc. Indian Acad. Sci.*, 1959, **50B**, 305—318).—Growth and yield of grain was optimum in media containing phosphate 5—10 and K 20 p.p.m. respectively. Higher levels of phosphate over 40 p.p.m. were toxic and reduced grain yield but no adverse effect resulted from high K levels. (16 references.)

E. G. BRICKELL.

Potato cultivation with moderately sound seed potatoes. A. J. Reestman (*Landbouwoorlichting*, 1960, **17**, 64—70).—Earlier trials indicated that inferior seed potatoes can best be utilised by planting more densely than usual. Stunted plants can be removed subsequently provided that there is a normal quota of sound

plants. The selection of the larger tubers for planting is not recommended. Leaf-production by the infected plants may be improved by extra N-manuring.

P. S. ARUP.

Effect of fertilisers on the chipping quality of freshly-harvested and stored Red River Valley potatoes. H. Findlen (*Amer. Potato J.*, 1960, **37**, 85—89).—Application of N (60—180 lb.), P (60—120 lb.) and K (60 lb. per acre) in various combinations had no effect on the quality of chips produced just after harvest or after 5 months' storage of the tubers at 4.4°. All treatments produced darker coloured chips after storage. Kennebec gave lighter coloured chips than did Irish Cobbler.

A. H. CORNFIELD.

Nutrient composition of White Rose potatoes during growth and after storage. M. Yamaguchi, J. W. Perdue and J. H. MacGillivray (*Amer. Potato J.*, 1960, **37**, 73—76).—The Ca, Fe, P, vitamins C, B₁, B₂, and niacin, wt. per tuber, water content, total sugars, starch, protein and calorific energy of tubers of White Rose potatoes during growth and after storage at two temp. for varying periods are presented.

A. H. CORNFIELD.

Effect of waxing potatoes on weight loss, shrivelling, decay and appearance. R. E. Hardenburg, H. Findlen and H. W. Hruschka (*Amer. Potato J.*, 1959, **36**, 434—443).—The application of clear or coloured wax emulsions to potatoes prior to storage in bags at 4.4° or 21.1° for 6 weeks was ineffective in reducing wt. loss, shrivelling, decay or otherwise in improving keeping quality. Red wax tended to improve the colour of red-skinned varieties.

A. H. CORNFIELD.

Non-destructive technique for detecting internal discolorations in potatoes. G. S. Birth (*Amer. Potato J.*, 1960, **37**, 53—60).—A spectrophotometer for recording spectral absorption curves of tubers is described. Hollow heart tubers had a higher transmittance at 710 m μ than had normal tubers. The method was 98% effective in detecting hollow heart in Irish Cobbler tubers and 81% effective in detecting internal discolorations due to hollow heart, decay, greening and blackspot in Katahdin tubers.

A. H. CORNFIELD.

Effect of harvest date and rate of nitrogen fertilisation on maturity, yield and chipping quality of potatoes. G. W. Hope, D. C. MacKay and L. R. Townsend (*Amer. Potato J.*, 1960, **37**, 28—33).—Maturity of potatoes (measured by degree of die-down of tops) decreased with increasing level of N fertilisation (80—240 lb. per acre). Chip colour improved as potatoes matured but declined with excessive levels of N. The highest yields giving acceptable chip quality occurred with 160 lb. of N per acre and in crops harvested when at least 80% of the leaves had died. There was a significant negative correlation between chip colour and both reducing sugars and amino-N in the tubers.

A. H. CORNFIELD.

Correction of leaf necrosis of potatoes with foliar and soil applications of potassium. W. M. Laughlin and C. H. Dearborn (*Amer. Potato J.*, 1960, **37**, 1—12).—Foliage sprays or soil applications of KCl reduced foliar necrosis and increased the yields of tubers. Max. benefits occurred with the application of 160 lb. of K₂O per acre. Foliar sprays of K were effective in reducing the incidence of brown sunken lesions of tubers only when K was also applied to the soil.

A. H. CORNFIELD.

Effect of time of planting on the occurrence of internal brown spot in the potato variety Arran Banner in Lebanon. A. A. Ahmadi, H. Mobarak and J. Osguthorpe (*Amer. Potato J.*, 1960, **37**, 23—27).—Planting in Feb. or March produced tubers having 40—78% internal brown spot at two locations over 2 years. Incidence of the disorder decreased in later plantings (min. with June plantings). Yields decreased with delay in planting. June plantings produced tubers of the best keeping quality.

A. H. CORNFIELD.

Interaction of chloride with sulphate and phosphate in the nutrition of potato plants. E. G. Corbett and H. W. Gausman (*Agron. J.*, 1960, **52**, 94—96).—The SO₄²⁻ content of the tops of potato plants grown in sand cultures decreased and that of Cl⁻ increased with increasing Cl⁻ concn. (0—600 p.p.m.) in the nutrient. PO₄³⁻ in the tops was highest at the lowest and highest levels of Cl⁻. Tuber- and root-SO₄²⁻ and -PO₄³⁻ were unaffected by level of Cl⁻. Addition of ³²P increased the uptake of SO₄²⁻ by the roots, whilst ³²S had no effect.

A. H. CORNFIELD.

Comparison of nitrogen sources for mid-season fertilisation of sugar-beet. R. S. Loomis, J. H. Brickley, F. E. Broadbent and G. F. Worker, jun. (*Agron. J.*, 1960, **52**, 97—101).—On a calcareous clay loam application of N (120 lb. per acre) as NH₃ (injected into soil), (NH₄)₂SO₄, urea, NH₄NO₃ or Ca(NO₃)₂ 110 days after sowing resulted in similar increases in yields of beet and sucrose per acre. Application of NH₃ in the irrigation water gave lower yield increases. On a non-calcareous loam (pH 7.6) none of the N sources applied as a side-dressing (80 lb. N per acre) 60 days after sowing had any effect on yields of beet or sucrose. Petiole NO₃⁻ level showed the

quickest increase and highest values where NO₃⁻ compounds were applied, followed in order by urea and NH₄ compounds.

A. H. CORNFIELD.

Change of forage seed quality under different simulated shipping conditions. T. M. Ching, H. L. Taylor and P. T. Rowell (*Agron. J.*, 1960, **52**, 37—40).—When stored in cloth bags at 32.2° and 90% R.H. six types of forage seed showed very poor germination and increased moisture content after a few weeks. Storage at 1.7° and 85% R.H. or at 21.1° and 66% R.H. resulted in little decline in viability after 10 weeks but some decline after 6 months. Fresh seed and seed with high original germination withstood the adverse conditions better than did one-year-old seed and seed of original poor viability. Storage in polyethylene bags maintained high viability even at 32.2° and 90% R.H.

A. H. CORNFIELD.

Nitrate content of herbage at different manurial levels. G. ap Griffith (*Nature, Lond.*, 1960, **185**, 627—628).—Grass sward was treated in April with 0—12 cwt. of (NH₄)₂SO₄ per acre. Determinations of NO₃⁻-N 6 weeks later showed detectable NO₃⁻ in the grass at the 4-cwt. level, increasing to 0.25—0.30% at the 12-cwt. level. The NO₃⁻ content was correlated with crude protein content. Forage containing more than 0.07% of NO₃⁻-N (dry wt. basis) is reported to be toxic to animals; this level may be reached at about 22% crude protein.

M. D. ANDERSON.

Effect of autumn application of nitrogenous fertiliser to irrigated pasture on production of grazing dairy cows. M. Freer and D. E. Tube (*J. Aust. Inst. agric. Sci.*, 1959, **25**, 306—308).—Although no significant differences in pasture yields resulted from applications of N (150 lb./acre as urea) the milk yields were significantly increased.

A. G. POLLARD.

Changes in soluble sugar contents of short rotation ryegrass during growth. J. S. Couchman (*J. Aust. Inst. agric. Sci.*, 1959, **25**, 315—319).—Sucrose contents of the grass mown at intervals during growth reached max. just before the formation of flower buds, decreasing subsequently. Variations in fructosan contents were similar. At the heading stage fructosans were the principal sugars and this stage is probably the best for cutting for silage.

A. G. POLLARD.

Nitrogen content of grass lignin. D. L. Whitehead and G. V. Quicke (*J. Sci. Fd Agric.*, 1960, **11**, 151—152).—Lignin from veld grass appears to contain N, partly in the form of -NCH₃ groups, not wholly removable by repeated purifications with dioxan. (11 references.)

E. M. J.

Distribution and condition of soil phosphate under old permanent pasture. B. Roscoe (*Plant & Soil*, 1960, **12**, 17—29).—Total and Morgan-extractable P to a depth of 36 in. is reported for a clay loam which had been under permanent pasture for 57 years and had received varying fertiliser treatments during this period. Both basic slag and superphosphate increased total soil-P, the greatest effect occurring in the 0—3 in. soil layer and declining with depth. Morgan-extractable P also decreased with depth and the highest values in the surface layer occurred where superphosphate or basic slag + lime had been applied. When the soils were cropped to wheat and oats in the two years following ploughing highest yields occurred where basic slag + lime had been applied.

A. H. CORNFIELD.

Mineral composition of herbage in relation to development of hypomagnesaemia in grazing cattle. J. A. F. Rook and M. Wood (*J. Sci. Fd Agric.*, 1960, **11**, 137—143).—The contents of inorg. constituents of herbage samples are considered in relation to the observed incidence of hypomagnesaemia. For a given sward in a given season, the severity of the disease in cows grazing plots given different fertiliser treatments, generally increased as the alkaline earth alkalinity (Ca + Mg - P, expressed as mequiv./100 g. silica-free herbage dry matter) of the herbage decreased. (17 references.)

E. M. J.

Influence of sod-seeded legumes on the nitrogen economy of grassland soils at Lismore, N.S.W. J. W. McGarity (*J. Aust. Inst. agric. Sci.*, 1959, **25**, 287—293).—Sowing clover over a *Paspalum* pasture results in a gain of 40—60 lb. of N per acre.

A. G. POLLARD.

Influence of temperature on the symbiotic nitrogen fixation of legumes. M. G. Mes (*Nature, Lond.*, 1959, **184**, 2032—2033).—Plants of *Vicia sativa*, *Pisum sativum* and *Lupinus luteus* grown in N-free nutrient solutions, from seeds inoculated with *Rhizobium*, showed lower N (mg./plant) and N (% dry wt.) when day temp. rose from 18°, 19° or 21° to 25° or 27°, and usually lower total N with increase of night temp. from 10° to 21° (although % N was often increased). In legume spp. normally growing in tropical or sub-tropical areas (e.g., velvet bean, groundnut, soya-bean), total N and % N increased with increasing day or night temp. Velvet bean and groundnut showed little N fixation with night temp. below 18°. With *Trifolium pratense*, the effects of temp. on N fixation were much less consistent, and of less importance than other factors.

M. D. ANDERSON.

Symbiotic nitrogen fixation in a grazed tropical grass-legume pasture. A. W. Moore (*Nature, Lond.*, 1960, **185**, 638).—After 5 years' growth of a pure grass stand (*Cynodon plectostachyum*) and of a mixture of this grass with the legume *Centrosema pubescens*, on a sandy soil in Nigeria, the N content of the top 12 in. of soil was 560 lb. per acre higher under the grass-legume mixture. N fixation by pasture legumes may thus be as extensive and important in tropical as in temperate regions. M. D. ANDERSON.

Internal breakdown in the crown of red clover. J. H. Graham, C. L. Rhykerd and R. C. Newton (*Plant Dis. Repr.*, 1960, **44**, 59—61).—Symptoms of an internal breakdown in the crown of red clover at many locations are described. Various organisms were isolated from affected crowns, but attempts to produce the condition artificially by inoculation of healthy crowns and roots with fungi, bacteria and ground tissue from affected plants were not successful. No macroscopic growth-deficiency symptoms were observed and there were no responses to additions of trace elements. A. H. CORNFIELD.

Response of lucerne to boron fertilisation of acid and calcareous soils in a greenhouse study. M. A. Norland and R. W. Starostka (*Agron. J.*, 1960, **52**, 33—35).—Application of borax (30 lb. per acre) to free calcareous soils in greenhouse tests had no effect on lucerne yields over six cuttings, although %B in the plant was usually increased at all cuttings. Lucerne grown in acid soils which had been limed showed B-deficiency symptoms and had low %B in the tissue. Application of borax (10—20 lb. per acre) eliminated B-deficiency symptoms, greatly increased %B in the tissue and increased yields of lucerne in some cases. A. H. CORNFIELD.

Evaluation of the maximum radius of the rosette as a growth index for subterranean clover. E. A. N. Greenwood and E. G. Hallsworth (*Plant & Soil*, 1960, **12**, 49—56).—The max. radius of the rosette (distance from the centre shoot to the base of the leaf furthest from it) of subterranean clover (Mt. Parker strain) had a correlation coeff. of 0.98—0.93 with dry matter yields over the early vegetative period, but this correlation coeff. decreased with the age of the plant. Providing the correlation coeff. between the two factors at harvest is >0.85, rosette radius may safely be used as an index of dry wt. at any period prior to harvest. A. H. CORNFIELD.

Storage of pineapple shoots in the dry season. C. Py (*Fruits d'outre mer*, 1960, **15**, 29—32).—Shoots were harvested at intervals throughout the dry season, stored and then planted in May. The loss in wt. of shoots during storage, and the height, leaf size and fruit production of plants grown from these shoots, was recorded. Shoots were stored more successfully when placed vertically in the shade than in the sun. Regular watering of shaded shoots had no significant effect. S. G. AYERST.

New diagnostic technique for micronutritional disorders of oranges. E. Giovannini and P. Fichera (*Ric. sci.*, 1960, **30**, 128—137).—The effect of trace elements on the activity of chloroplast materials extracted from orange leaves has been studied by a bioelectric potential method based on the reduction of Fe³⁺ in aq. solution under the influence of chloroplast and light. The method provides a basis for studying micro-nutritional disorders by observing the effect of trace elements on the potential curve. L. A. O'NEILL.

Effect of soil salinity on growth of broad beans, *Vicia faba*. A. D. Ayres and D. L. Everhard (*Agron. J.*, 1960, **52**, 110—111).—Increasing soil salinity by addition of NaCl + CaCl₂ decreased the dry matter yields of broad bean plants. A 50% reduction in yield occurred with salinity levels corresponding to 6—7 millimhos in the saturation extract, indicating that the plant has a moderate tolerance to salinity. The % of Na, Ca and Cl in the plant increased considerably with level of salinity, that of Mg increased slightly, whilst K decreased. A. H. CORNFIELD.

Abnormal growth and development in pea resulting from exposure to adverse conditions during germination. A. M. M. Berric (*Nature, Lond.*, 1960, **185**, 626—627).—Pea seeds were held for 48 hr. at 20° and in darkness (i) in an atm. containing 20% of CO₂, (ii) in a large vol. of tap water, (iii) in running tap water. All treated seeds subsequently produced plants with fewer and smaller flowers than the controls, the effects being more marked with treatments (i) and (ii) than with (iii). Gross abnormalities of development were commonest in (ii). M. D. ANDERSON.

Damaging effect of drying on *Vicia faba* seeds. W. Klingmüller and G. R. Lane (*Nature, Lond.*, 1960, **185**, 699—700).—When seeds of *Vicia faba* were dried over CaCl₂, water contents fell from ~12% for beans stored at room temp. to 7.5% after 4 weeks over CaCl₂. Germination % was progressively reduced by drying, as was the

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rate of growth of roots and shoots on germination. The effect of drying alone must be taken into account in radiobiological work on seeds. M. D. ANDERSON.

Effects of several patterns of defoliation on fruit production of the tomato plant (*Lycopersicon esculentum*, Mill.) with reference to possible paths of carbohydrate movement from leaves to fruit. K. K. Brubaker (*Dissert. Abstr.*, 1959, **20**, 1940).—The rate at which tomato fruits matured was not affected by removal of leaves. The dry wt. of the fruit was often reduced by removal of leaves, but there was usually no corresponding reduction in the size or fresh wt. of the fruit. In general, leaf area seemed to have more influence on the dry wt. of the fruit than the position of the leaves removed. M. D. ANDERSON.

pC value of soils and its effect on the tomato crop. R. E. Butters (*J. Sci. Fd Agric.*, 1960, **11**, 202—212).—Blotchy ripening of tomatoes can be reduced without loss of yield on soils that contain little or no gypsum if the pC of the 1 : 2½ water-extract is maintained between 3.0 and 3.2. Excessive transpiration of plants on drier soils increases blotchy ripening. On soils with high gypsum levels, use of the saturation extract to determine the salt concn. gives good correlation with plant growth; the 1 : 2½ extract is unsatisfactory. Gypsum has little, if any, effect on plant growth even when added in quantity sufficient to saturate the 1 : 2½ extract. In the 1 : 2½ extract gypsum gives a low pC reading but its effect in the soil solution is small. E. M. J.

Nitrogen balance of buckwheat. H. Roschack (*Z. PflErnähr. Ding.*, 1960, **88**, 18—35).—The N balance of buckwheat/nutrient solution systems containing antibiotics which destroy micro-organisms, able to convert atm. N into plant-available N, show a slight gain in N, i.e., buckwheat can assimilate atm. N. M. LONG.

Determination of the phosphorus status of *Hevea brasiliensis* by bark analysis. H. W. van der Marel (*Plant & Soil*, 1960, **12**, 5—16).—The P content of *Hevea* bark gave a better indication of P availability than did chemical methods of assessing soil P status or leaf analysis. Bark-P values were not subject to the variations found in leaves due to time of sampling. Bark-P values were correlated with soil org. P values in five soil types. A. H. CORNFIELD.

Yield response of cotton in the Sudan Gezira to DDT spray. R. J. V. Joyce (*Bull. ent. Res.*, 1959, **50**, 567—594).—Spraying cotton with DDT at 6—10 weeks old increased yields to extents which varied with season and site and which tended to be greater where N fertilisers had been applied. A. G. POLLARD.

Effect of spacing of soya-bean plants between and within rows on yield and its components. W. F. Lehman and J. W. Lambert (*Agron. J.*, 1960, **52**, 84—86).—The effects of space between rows (20 in. or 40 in.) and within rows (4—24 plants per ft. of row) on yield components of two varieties of soya-bean at two locations were studied. Seed yields tended to be higher with the narrower spacing between rows. Seed wt. differences due to between-row spacing varied with variety and location. Seed wt. increased with increasing within-row spacing at only one location. No. of seeds per pod increased with spacing between plants. Seeds per plant and pods per plant increased markedly as spacing was increased. A. H. CORNFIELD.

Radiation-induced leaf spot-resistant mutants in the groundnut, *Arachis hypogaea*, L. W. E. Cooper and W. C. Gregory (*Agron. J.*, 1960, **52**, 1—4).—Groundnut seed irradiated with 10,000—18,500 roentgens X-rays produced X₁ and X₂ progenies with both decreased and increased resistance to leaf spot, *Cercospora arachidicola*, Hori and *C. personata*, (B. and C.) Ell. and Ev. Selected entries compared in the following generations showed highly significant differences in no. and relative frequency of specific leaf spot lesions, no. of leaves not defoliated and in entry defoliation × disease control. With two exceptions selections carried into X₁, X₂ and X₃ were stable for the defoliation characteristics. A. H. CORNFIELD.

Pest Control

Incubation of soil and root samples in polyethylene plastic for improved recovery of nematodes. A. C. Tarjan (*Plant Dis. Repr.*, 1960, **44**, 31—35).—Incubating infested soil and root samples in sealed polyethylene plastic bags for 6—7 days at 23.9° substantially increased the yields of burrowing nematodes. Lower and higher incubation temp. gave lower recoveries of nematodes. A. H. CORNFIELD.

Separation of nematodes from soil by a modified Baermann funnel technique. J. T. Walker and J. D. Wilson (*Plant Dis. Repr.*, 1960, **44**, 94—97).—The technique is described. A. H. CORNFIELD.

Action of the systemic insecticide fluoroacetamide on certain aphids and on *Pieris brassicae*. W. A. L. David and B. O. C. Gardiner (*Bull. ent. Res.*, 1959, **50**, 25—38).—In general fluoroacetamide (I) was as effective as Na fluoroacetate and superior to schradan in tests against *Aphis fabae*, *Brevicoryne brassicae* and *Myzus persicae* by dipping, by soil application and systemically. The hatching of eggs of *Pieris brassicae* was not prevented by I but newly hatching larvae were killed. A. G. POLLARD.

Persistence of larvicides against *Culicoides*: interpretation of population changes in untreated plots. D. S. Kettle, R. H. Parish and J. Parish (*Bull. ent. Res.*, 1959, **50**, 63—80).—High persistence in soil of DDT and dieldrin used against *Culicoides impunctatus* is demonstrated. BHC was less effective. The nature of the formulation and, probably, variations in soil water levels affected the results obtained. A. G. POLLARD.

Fungicidal activity of δ -isomer and of mixtures of δ - and γ -isomers of hexachlorocyclohexane. K. A. Gar, N. V. Evtseva and E. I. Andreeva (*Dokl. Akad. Nauk SSSR*, 1959, **127**, 1290—1293).—In tests on *Fusarium oxysporum* and *Diplodia zaeae*, (Schw.) Lev., the γ -isomer suppressed growth of the latter fungus only, while the δ -isomer was effective against both and also against *Botrytis* sp., one *Penicillium* species, and yeasts. The fungicidal activity of the two isomers was greatest at 0.01% concn. At the higher concn. the growth of the micro-organisms was accelerated, and the formation of metal-chelate complexes was suspected. In tests on the seeds of wheat, flax and maize contaminated with various fungi, the effect of the prep. containing 12% of the δ - or γ -isomer, or their 1:1 mixture was compared with that of the Hg-containing fungicides. Both isomers were found suitable for seed protection, and results in % germination and the fungicidal action of the δ - γ isomer mixture exceeded those of the usual fungicides. A. L. GROCHOWSKI.

Regional seed-treatment tests for the control of seed-borne and soil-borne common bunt of winter wheat in the Pacific Northwest, 1959. L. H. Purdy (*Plant Dis. Repr.*, 1960, **44**, 25—28).—Results obtained with 21 materials at seven locations are reported. A. H. CORNFIELD.

Screening of potato fungicides in 1959. L. C. Callbeck (*Plant Dis. Repr.*, 1960, **44**, 68—70).—Of 12 materials tested, LO-1499 (NH_4 ethylenebis-dithiocarbamate) was the most effective in reducing defoliation and tuber rot due to late blight and in increasing yields. A. H. CORNFIELD.

Stem streaks of potatoes. D. B. Robinson, G. D. Easton and R. H. Larson (*Amer. Potato J.*, 1960, **37**, 67—72).—Stem streaking due to infection by *Phytophthora infestans*, *Verticillium albo-atrum*, virus Y and Mn toxicity are described and illustrated. There were varietal differences in susceptibility to the different forms of streak. High levels of applied Mn increased the intensity of *Verticillium* streak symptoms in Irish Cobbler, particularly at high soil moisture levels, even though there were no significant differences in Mn level between *Verticillium*-infected plants and healthy plants. Non-infected plants did not show streak symptoms even at high levels of applied Mn. A. H. CORNFIELD.

Eradication of bacterial ring rot, *Corynebacterium sepedonicum* from seed stocks of a potato breeding programme. R. B. O'Keefe and H. O. Werner (*Amer. Potato J.*, 1959, **36**, 427—433).—At two locations the Gram-stain and u.v. light methods were 95.3–99.9% effective in detecting ring rot freedom in hill selections. Despite the use of CuSO_4 and formalin as disinfectants, new or disinfected baskets and sacks and special machinery to reduce the likelihood of spreading infection during harvesting and grading, ring rot continued and spread for 2 years after the initial infection, but since then has not been detected for 4 years. A. H. CORNFIELD.

Potato scab control on organic soils. I. Initial response to pentachloronitrobenzene. H. T. Erickson (*Amer. Potato J.*, 1960, **37**, 18—22).—Incidence of potato scab in potatoes decreased with rate of application of pentachloronitrobenzene (100—300 lb. active material per acre, applied as a 20% dust disced into the soil one day prior to planting). Residual activity was apparent the year following the application of the higher rates. A. H. CORNFIELD.

Control of beet yellows. M. Heuver (*Landbouwoorlichting*, 1960, **17**, 81—87).—Satisfactory control of the aphid vectors can be achieved by spraying with Endothion, Fosfamidon, demeton or demeton-methyl (thiol-isomer). Increases in sugar yields after one or three (fortnightly) sprayings were 38 or 44%, respectively. On sandy soil, the increase after two sprayings was 23%. P. S. ARUP.

Influence of temperature on growth and nodulation of white clover infected with bean yellow mosaic virus. J. H. Smith and P. B. Gibson (*Agron. J.*, 1960, **52**, 5—7).—Max. growth and most conspicuous bean yellow mosaic virus symptoms of white clover grown

at temp. ranging from 10° to 30° occurred at 16.7°. At 10° the virus had no visible influence on growth. Root growth was max. at 10°. Rhizobia inoculation increased clover yields at all temp., whilst virus infection decreased the beneficial effects of rhizobia at 16.7° and higher temp. A. H. CORNFIELD.

Post-harvest fungicide treatments for the reduction of decay in Anjou pears. C. F. Pierson (*Plant Dis. Repr.*, 1960, **44**, 64—65).—Dipping Anjou pears in 0.12% captan, 0.12% Phaltan or 0.6% Na *o*-phenylphenate prior to storage at -0.5° for 150 days resulted in moderately good control of decay. A. H. CORNFIELD.

Control of bacterial spot, *Xanthomonas pruni*, of peach. U. L. Diener and C. C. Carlton (*Plant Dis. Repr.*, 1960, **44**, 136—138).—Application of 65% Dodine (n-dodecylguanidine acetate, 1.5 lb.) + captan (1 lb. per 100 gal.) in nine sprays during April—June gave excellent control of bacterial spot of peach. Sprays of captan or S (3 lb. per 100 gal.) were not quite as effective. Dodine alone or in combination with S was relatively ineffective. A. H. CORNFIELD.

Control of rusty spot of peach. R. H. Daines, C. M. Haenseler, E. Brennan and I. Leone (*Plant Dis. Repr.*, 1960, **44**, 20—22).—The best control of rusty spot of peach, probably due to a powdery mildew, was obtained by application of wettable S (5 lb. per 100 gal.) in four approx. weekly applications during May and early June. A. H. CORNFIELD.

Control of peach scab, *Cladosporium carpophilum*, Thum. C. H. Graves, jun., and B. C. Hurt, jun. (*Plant Dis. Repr.*, 1960, **44**, 129—131).—Application of Puratised Apple Spray (HgPh monoethanolammonium acetate, 2.5 pints per 100 gal.) during dormancy (March 2) followed by regular spraying with captan gave as effective control of peach scab when the March and early April captan sprays were eliminated as when the complete captan spray schedule was given. A. H. CORNFIELD.

Control of "death-bud" in sweet cherry. H. R. Cameron (*Plant Dis. Repr.*, 1960, **44**, 139—143).—A death of dormant buds in sweet cherry was ascribed to a strain of *Pseudomonas syringae*. Of a number of materials tested the best control was given by Puratised Agricultural Spray (HgPh monoethanolammonium acetate, 1 pint per 100 gal.) and Bordeaux mixture. A. H. CORNFIELD.

Natural infection of strawberry (*Fragaria* sp.) with tobacco-mosaic virus. P. Cornuet and J.-C. Morand (*C. R. Acad. Sci., Paris*, 1960, **250**, 1583—1584).—By extracting the leaves with a $(\text{NH}_4)_2\text{SO}_4$ -KCN solution which secures the removal of tannins, filtering and dissolving the ppt. in Sørensen phosphate buffer, stable aq. extracts were obtained from three varieties of *Fragaria* infected with tobacco-mosaic virus. Effects of inoculation of cultivated and wild strawberries with strains isolated from tobacco were studied. A. J. B.

Control of clubroot of cabbage in North Carolina. N. N. Winstead and H. R. Garriss (*Plant Dis. Repr.*, 1960, **44**, 14—18).—Application of pentachloronitrobenzene (2—6 lb. per 100 gal. of transplant water, 0.5 pints per plant) effectively controlled clubroot in a quick-growing, but not in a slow-growing cabbage variety. Treatment of the soil with methyl bromide (1—3 lb. per 100 sq. ft.) gave excellent control of both clubroot and wirestem disease, due to *Rhizoctonia solani*, Kuehn. A. H. CORNFIELD.

Effect of insecticide-fungicide combinations on emergence of peas and growth of damping-off fungi. L. T. Richardson (*Plant Dis. Repr.*, 1960, **44**, 104—108).—In soil infected with *Pythium ultimum* and *Rhizoctonia solani* pea seeds treated with insecticides (aldrin, dieldrin or γ - $\text{C}_6\text{H}_4\text{Cl}_2$) alone were more vulnerable to pre-emergence damping-off than were untreated seed, whereas seed treated with insecticide in combination with fungicides (thiram, captan or chlor-anil) showed less damping-off than when fungicides alone were used. In the absence of the pathogens none of the treatments had any effect on emergence. In culture media inoculated with the pathogens the insecticides, particularly γ - $\text{C}_6\text{H}_4\text{Cl}_2$, retarded mycelial growth, whilst in combination they added to the inhibitory effect of the fungicides. A. H. CORNFIELD.

Control of cone rust, *Cronartium strobilinum*, of slash pine. O. C. Maloy and F. R. Matthews (*Plant Dis. Repr.*, 1960, **44**, 36—39).—A high incidence of cone rust of slash pine occurred in the central and western portions of north Florida. Live oak, *Quercus virginiana*, was present in most of the locations where rust infection was heavy, indicating that it is an important alternate host for the disease. Ferbam (2 lb. per 100 gal.) reduced infection when applied to strobili prior to the stage at which pollination is completed. A. H. CORNFIELD.

Heat treatment for control of mosaic disease, due to Marmor rosae, H., of rose. F. O. Holmes (*Plant Dis. Repr.*, 1960, **44**, 46—47).—Small infected plants were held in an incubator for 4—10 weeks at temp. ranging from 33.5 to 34.0° for 1—4 weeks and at 36° for the

rest of the time. After removal from the incubator new foliage showed no symptoms during 40 weeks' growth.

A. H. CORNFIELD.

The coconut pest, *Pseudotheraptus wayi*, Brown, in Zanzibar. II. Yields of coconuts in relation to damage caused by the insect. III. A selective residual insecticidal formulation and its effects on the ecology of the insect. F. L. Vanderplank (*Bull. ent. Res.*, 1959, 50, 135—149, 151—164).—III. Customary prep. of BHC controlled *P. wayi* but also destroyed predatory ants. A formulation of DDT with a coumarone-indene resin in petroleum oil killed *P. wayi* walking over it but did not destroy the larger predatory ants, e.g., *Oecophylla longinoda*.

A. G. POLLARD.

Possible control of the mealy bug vectors of cacao swollen-shoot virus by trunk implantation with dimefox. A. D. Hanna, W. Heatherington, H. R. Mapother and R. Wickens (*Bull. ent. Res.*, 1959, 40, 209—225).—Promising results were obtained in controlling the vectors by implantation of dimefox in holes drilled in the trunks at ground level.

A. G. POLLARD.

Chemical control of cacao Mirids, *Distantiella theobroma* (Dist.) and *Sahlbergella singularis*, Hagl. F. Raw (*Bull. ent. Res.*, 1959, 50, 13—23).—Cacao beans were sprayed with DDT or BHC and the Mirids were placed on the dried residue. BHC had the more rapid "knock-down" action and the higher toxicity. Fumigant effects from sprayed leaves were considerable but short-lived with BHC and negligible with DDT. DDT residues retained toxicity to nymphs for about 3 weeks and BHC for 2—3 days only.

A. G. POLLARD.

Herbicides. III. 2,4-Disubstituted-6-chloro-1,3,5-triazines. H. Koopman and J. Daams (*Rec. Trav. chim. Pays-Bas*, 1960, 79, 83—89).—Several 6-chloro-1,3,5 triazines 2,4 disubstituted by alkyl-oxy, -thio or -amino are prepared by action of alcohol, thiol or amine on the 2-substituted compound. The herbicidal activity is less by watering the roots than by spraying the leaves. Compounds with alkoxy or alkylthio in the 2- and 4-positions cause rapid leaf damage, but alkylamino-substitution decreases the activity.

J. A. C. ABSTR.

Retention and effect of 2,4-dichlorophenoxyacetic acid (2,4-D) sprays on winter wheat. H. D. Woofter (*Dissert. Abstr.*, 1959, 20, 1949—1951).—Retention of 2,4-D sprays on wheat in the field was followed by colorimetric determination of the water-sol. dye Anthraquinone Blue, incorporated in the spray. Retention increased at first with age of plant at spraying, but decreased slightly from the stage of formation of the ear. Retention was lower with 12 gal. of spray per acre than with 3 or 6 gal. Retention increased with the amount of 2,4-D applied, and was affected by formulation, the isopropyl ester giving the highest retention, with the triethanolamine salt + wetting agent next in order. The plants were most resistant to damage by the spray between full tillering and jointing, and most susceptible at the early-head stage. Damage included diminution of kernel wt., and lessened germination %. Amounts of 2,4-D sufficient to control most broad-leaved weeds ($\frac{1}{4}$ — $\frac{1}{2}$ lb. per acre) can safely be applied to winter wheat at the fully-tillered stage.

M. D. ANDERSON.

Effect of 1,2,4,5-tetrachlorobenzene on the germination and seedling vigour of barley, oats and wheat. O. A. Ameen, A. D. Day and K. C. Hamilton (*Agron. J.*, 1960, 52, 87—89).—The effects of 1,2,4,5-tetrachlorobenzene (I) (1.7—135 lb. per acre) on growth of cereals sown on four soil types from 1 to 125 days after application of I are reported. The detrimental effect of I was most pronounced on sandy soil and decreased with fineness of soil texture and with length of time between application of I and sowing of the cereals. No injury to the plants occurred when the seed was sown 125 days after application of even the heaviest application of I.

A. H. CORNFIELD.

Instability in wheat and barley produced by application of dalapon. C. A. Suneson and L. G. Jones (*Agron. J.*, 1960, 52, 120—121).—Application of dalapon for the control of wild oats resulted in the production of variously malformed oat and barley plants which had been sown before the herbicide had decomposed completely. These abnormalities, together with new diversities, were carried through to four generations.

A. H. CORNFIELD.

Mistletoe control on a large scale. A. G. Brown (*J. Aust. Inst. agric. Sci.*, 1959, 25, 282—286).—Details of a system for calculating the dosage of 2,4-D to be applied by injection (axe-cut), using a drenching gun, into trunks of infested trees.

A. G. POLLARD.

Control of fungus organisms. American Cyanamid Co. (Inventor: G. Lamb) (B.P. 812,492, 28.1.57).—A dry composition, for use in the control of fungus organisms which attack living plants (e.g., *Monilinia fructicola*, *Stemphylium sarcinaefforme*, *Alternaria solani*, *Colletotrichum lagenarium*, *Phytophthora infestans*, etc.), comprises an acid salt of dodecylguanidine (especially the acetate) and an inert

carrier (talc, kaolin, methylcellulose, bentonite, fuller's earth, Attapulgitic clay, pumice, SiO₂, a silicate or CaCO₃). The composition may also be used in the form of an aq. suspension containing a wetting agent.

F. R. BASFORD.

Esters of [5-acyl-8-hydroxyquinolines]. J. R. Geigy A.-G. (B.P. 812,177, 15.6.56. Switz., 16.6.55).—A conventional method is described for the prep. of the esters (which may have a 2-methyl substituent, and acyl is benzoyl or alkanoyl of >5 C) useful as bacteriostats (active against a number of pathogenic bacteria) and fungicides (*Tilletia tritici*, *Fusarium culmorum*, *Ctenomyces interdigitalis*). One example given is a formulation containing ethyl 5-acetylquinol-8-yl carbonate, useful for disinfection of bedding earth and for dusting bulbs and tubers.

J. A. C. ABSTR.

Polymeric phosphate esters. Union Carbide Corp., Asse of W. N. Lanham (B.P. 812,390, 5.6.56. U.S., 6.6.55).—Compounds useful, *inter alia*, as insecticides comprise dialkyl (or diaryl, dialkaryl or diaralkyl) polyepoxyalkyl (2—4 C) esters of phosphoric acid, the prep. of which is described.

J. A. C. ABSTR.

Substituted 2-mercaptoethyl thiophosphoric acid dialkylates. Farbenfabriken Bayer A.-G. (B.P. 812,065, 29.7.55. Ger., 2.8.54).—The compounds, useful as pesticides, are obtained by treating a (2-bromoethyl) thiophosphoric acid dialkyl ester with a linear aliphatic, an aromatic or an aromatic-heterocyclic-fused ring compound containing SH (or a salt thereof) in neutral or alkaline medium. *OO*-diethyl *S*-2-bromoethyl phosphorothiolate is converted into *OO*-diethyl *S*-(2-ethylthioethyl) phosphorothiolate, b.p. 82°/0.01 mm.

F. R. BASFORD.

Esters of monothiophosphoric acid and insecticidal compositions containing them. Sandoz Ltd. (B.P. 812,369, 30.8.55. Switz., 1.9.54).—Compounds OR(OR')·PS·O·CHR''·CH₂·SR''' (R, R' and R'' are hydrocarbon radicals of 1—3 C; R''' is alkoxyethyl of 2—4 C), useful as insecticides, are obtained by interaction of OR(OR')·PSX with OM·CHR''·CH₂·SR''' (X is halogen; M is H or alkali metal), preferably at 50—150° in presence of acid-binding agent in an inert diluent. Details are given for the prep. of *Me*₂ 3-methoxy-1-ethylthio-prop-2-yl thionophosphate, b.p. 128—132°/0.5 mm., *n*_D²⁰ 1.4945.

F. R. BASFORD.

Odour removal and stabilisation of phosphate-containing pesticides. American Cyanamid Co. (B.P. 812,182, 13.7.56. U.S., 12.8.55).—A pesticide OR(OR')·PS·S·CH(CH₂·CO₂R'')·CO₂R''' (R and R' are aliphatic or aromatic radicals; R'' and R''' are H, aliphatic or aromatic radicals) is deodorised and stabilised by treatment with 0.01—1 wt.-% of a peroxide, e.g., Bu₂O₂, cumene or pinene hydroperoxide, Bu^t pentamethylethyl peroxide, or H₂O₂. Thus, technical *OO*-dimethyl *S*-1,2-diethoxycarbonyl ethyl phosphorothiothionate (1000) is stirred with cumene hydroperoxide (0.5 pt.) during 15 min. at 25°, to give a premium grade product.

F. R. BASFORD.

New dithiophosphoric acid esters and insecticidal preparations containing them. Sandoz Ltd. (B.P. 812,119, 28.11.57. Switz., 12.12.56).—Compounds (OR)₂PS·S·CH₂·S·CH₂·OR', useful as insecticides and seed disinfectants (especially for the treatment of cotton seed), are obtained by treating (OR)₂PS₂H with S(CH₂·OR')₂ at 20—150° (R and R' are alkyl of 1—4 C). *OO*-Dimethyl *S*-(methoxyethylthio)methyl thiothiophosphate, b.p. 100—102°/0.06 mm. is prepared. This (50), compounded with an emulsifying agent consisting of an alkylpolyglycol ether or an alkylphenylpolyglycol ether, e.g., iso-octylphenylheptaglycol ether (50 pt.), followed by dilution with water to 0.05% of ester, affords an aq. emulsion which is 100% effective in killing aphids on young apple trees.

F. R. BASFORD.

Phosphonic and thiophosphonic acid derivatives. Farbenfabriken Bayer A.-G. (B.P. 812,530, 22.8.57. Ger., 23.8.56 and 18.7.57).—Compounds R·SO_n·R'·PXR''·OR''' are obtained by treating R·SO_n·R'·OH (or a salt thereof) with (OR''')·PXR''·Cl, e.g., at 0—100° in an inert solvent, in presence of acid-binding agent (if desired). The products are useful as insecticides and compositions containing them for this purpose are also claimed (R and R' are alkyl or aryl; R' is aryl; R'' is alkyl of 1—4 C; *n* is 0—2; X is O or S). In an example, a description is given of the prep. of *Et p*-methylthiophenyl methylphosphonate, m.p. 90°/0.01 mm., in 71.5% yield. This is 100% lethal against Colorado beetles, black bean aphids and eggs of spider mites at respective concn. of 0.01, 0.001 and 0.0001.

F. R. BASFORD.

Compositions for combating nematodes. Farbenfabriken Bayer A.-G. (B.P. 812,512, 10.4.57. Ger., 12.4.56).—The active ingredient of the composition is a benzothiazole optionally substituted in the 2-position by alkyl of 1—5 C, halogen, NO₂, NH₂ or hydroxyalkyl of 1—5 C, and in the benzene nucleus by alkyl of 1—5 C, NO₂, halogen, SO₂H (or a salt form thereof) or NH₂. Thus, commercial emulsifier, e.g., polyglycol ether of benzylhydroxydiphenyl, is added

(10 wt.-%) to a solution of 2-chlorobenzthiazole in dimethylformamide, then the mixture is diluted with water to 0.01% of active ingredient, to provide a solution which is 100% effective against a variety of nematodes (*Meloidogyne* sp., *Ditylenchus dipsaci*, *Aphelenchoides ritzemabosi* and *Heterodera schachtii*).

F. R. BASFORD.

Soil fumigant composition. Dow Chemical Co. (B.P. 812,677, 28.10.57. U.S., 29.11.56).—A fumigant composition, especially useful against nematodes and wireworms, comprises a mixture of 1,2-dibromoethane (1—2) and 1,3-dichloropropane (3—9 vol.), preferably compounded with a diluent (petroleum hydrocarbon distillate, b.p. <205°, flash point >25°) or dispersed on or in a granular or finely divided solid carrier.

F. R. BASFORD.

Cycloalkyl derivatives of urea and herbicidal agents. Badische Anilin & Soda-Fabrik A.-G. (B.P. 812,120, 6.12.57. Ger., 14.12.56).—Compounds $[CH_2]_n \cdot CH \cdot NH \cdot CO \cdot NRR'$ are claimed (R and R' are alkyl, cycloalkyl, aryl, or together with N form a heterocyclic radical which may optionally include O; n is 6 or 7). The products are characterised by herbicidal activity. 1-Cyclo-octyl-3,3-dimethylurea, m.p. 137—137.5°, is prepared, which (2.5) in water (1200 kg.) when applied to a 1-hectare greenhouse containing *Sinapis alba*, *Avena sativa*, *Poa annua*, *Galium aparine*, *Galinsoga parviflora* or *Polygonum persicaria*, causes complete withering thereof within 10 days.

F. R. BASFORD.

Thio- and dithio-carbonic acid esters. Farbenfabriken Bayer A.-G. (B.P. 812,480, 6.12.56. Ger., 6.12.55).—Compounds, characterised by outstanding fungicidal properties without phytotoxic effect, comprise esters obtained by interaction of a cyclic oximide with R:C(X):YR' (R is halogen; X and Y are S or either X or Y is O; R' is alkyl or aryl). The prep. of N-(thionocarboxy)phthalimide, m.p. 118—119°, in 59.8% yield, is described.

F. R. BASFORD.

Animal Husbandry

Ruminant digestion study techniques. I. Cotton loops as a measure of cellulose digestion. II. Preservation of faeces to prevent nitrogen and dry matter losses. E. H. Haynes (*Dissert. Abstr.*, 1959, 20, 1934).—The rate of breakdown of the cellulose of cotton threads suspended in the rumen of fistulated steers was a sufficiently precise measure of the effects of variables on the digestibility of cellulose. Varying the ratio of hay to grain in the diet did not affect the rate of digestion of cellulose. The rate did not increase with time after feeding; it was fastest with shortest preliminary feeding periods. Extreme day-to-day fluctuations in the rate were observed. N was lost from stored faeces, however preserved. Drying fresh faeces resulted in losses of 7.5% of the dry matter and 3.6% of the protein content.

M. D. ANDERSON.

Development of an in-vivo artificial rumen. G. W. Teresa (*Dissert. Abstr.*, 1959, 20, 1972).—Rumen digestion was studied by placing within the rumen of a fistulated animal a porcelain test-tube, fitted with a frothing tube and gas escape mechanism, and containing substrates and inocula. The test-tube was permeable to volatile fatty acids and certain dyes; concn. of acids in the tube after 40 hr. was about 40% of that in the rumen itself. Continuous digestion of cellulose occurred in the tube, reaching 88%, and results were reproducible. Of the cellulose of first-cutting lucerne, 76% was digested, compared with 50% on the second cutting; the difference is attributed to lignification in the older material.

M. D. ANDERSON.

Continuous culture as a method for studying rumen fermentation. D. G. Stewart (*Dissert. Abstr.*, 1959, 20, 1937—1938).—The continuous-culture process is suitable for study of rumen fermentation *in vitro*. Changes in pH, rates of production and proportions of volatile fatty acids, no. of coliform and amylolytic organisms, and rates of growth of protozoa are examined.

A. M. SPRATT.

Use of forage and faecal protein as indicators of forage digestibility. J. A. Holter (*Dissert. Abstr.*, 1959, 20, 1934—1935).—The apparent digestibility of protein in forages was highly correlated with crude protein content. Sheep appear to digest the protein of low-protein forages more efficiently than do cattle. Correlations between contents of crude and of digestible protein were high. Various methods of predicting digestible protein values were examined. Calculation from the protein content of the faeces is a useful method. Separate equations are needed for fresh forages, dry forages and silages.

M. D. ANDERSON.

Nutritive value of maize and soya-beans as influenced by soil treatment. B. E. Rutherford and K. M. Pretty (*Agron. J.*, 1960, 52, 27—29).—Maize and soya-beans were grown on a sandy loam treated with lime, P and K in a complete factorial experiment. Wt. gains and N digestibility of rats receiving diets containing the seed were not significantly different between soil treatments. The soil lime

treatment tended to depress the rat-bone P%. Even though soil P treatments increased seed P% considerably, these treatments had little effect on rat-bone P%.

A. H. CORNFIELD.

Physiological effects of gossypol and cotton-seed pigment glands. E. Eagle (*J. Amer. Oil Chem. Soc.*, 1960, 37, 40—43).—Extraction methods and toxicity to laboratory mammals are reviewed. (38 references.)

P. M. KINGSTON.

Non-medical uses of antibiotics. III. Animal nutrition. R. Levin (*Chem. Prod.*, 1960, 23, 218—220).—A brief review dealing with chicks, turkeys, calves and pigs. Hypotheses explaining the beneficial effects are summarised. The "sub-clinical infective state" is specially stressed; with improved hygiene stock is healthier and can grow at optimal rates without recourse to antibiotics.

C. V.

Effect of feeding Aureomycin to Harijana calves on growth, body measurement, blood composition and parasitic infection. D. N. Mullick (*Indian J. Dairy Sci.*, 1959, 12, 100—105).—Aureomycin fed to calves, 14—17 weeks old, at the rate of 10, 30 or 90 mg. per 100 lb. live wt. for 13 weeks had no significant effect on growth rate, feed consumption, heart girth, leucocyte count or haemoglobin concn. There were significant reductions in helminthic egg counts in the faeces of calves fed the antibiotic and these calves had a good general appearance and were free from calldow diseases. (12 references.)

S. G. AYERST.

Use of reserpine as supplement to fodder for farm animals. H. Schneebberger and A. Schürch (*Mitt. Lebensm. Hyg., Bern*, 1959, 50, 523—532).—The supplement (1.25 mg. per kg. of feed) administered to fowls during the whole period of growth (12 weeks) or during the last 6 weeks, results in no significant change in carcass-wt., and is without effect on the incidence of perosis, feather-picking or heat-stress. (17 references.)

P. S. ARUP.

Critical features of good dairy feeding experiments. H. L. Lucas (*J. Dairy Sci.*, 1960, 43, 193—212).—The factors to be considered in designing dairy feeding experiments are critically surveyed. (44 references.)

S. C. JOLLY.

Net energy of blackstrap molasses for lactating dairy cows. G. P. Loigreen and K. K. Otagaki (*J. Dairy Sci.*, 1960, 43, 220—230).—When fed at the rates of 10 and 30% of the total ration, the net energy of cane or blackstrap molasses was 68.1 and 23.1 g.-cal. per 100 lb. respectively, compared with 54.2 g.-cal. for the basal ration of pineapple bran, soya-bean meal, dicalcium phosphate and trace-mineralised salt; the lower energy value at the higher rate of feeding was not due to increased faecal loss. The higher rate significantly decreased butterfat and solids-not-fat in the milk, resulting in a lowering of the energy value per unit of milk produced; the flavour of the milk was impaired, apparently due to increased acidity. None of these defects occurred with the lower rate of feeding. The caloric value of the milk solids was closely related to the fat content (X) of the milk, and is described by the equation: $Y = 4.516 + 0.321X$, where Y = the kg.-cal. of energy per g. of milk solids.

S. C. JOLLY.

Effect of milking intervals on the rate of milk and fat secretion. G. H. Schmidt (*J. Dairy Sci.*, 1960, 43, 213—219).—The rates of secretion of milk, fat and solids-not-fat were linear with milking intervals of 4, 8 and 12 hr., but with intervals of 16 and 20 hr. rates were significantly lower. Butterfat % was almost the same for the various intervals, except for the 4-hr. interval when the % was inexplicably high; the solids-not-fat contents were the same throughout. Preceding long intervals decreased milk secretion during 12-hr. intervals, but markedly increased butterfat %.

S. C. JOLLY.

Quantitative determination of free oestrone, 17 α -oestradiol and 17 β -oestradiol in bovine foetal cotyledons. E. I. Veenhuizen, R. E. Erb and J. Gorski (*J. Dairy Sci.*, 1960, 43, 270—277).—A method, based on partition extraction, paper-chromatographic separation and fluorimetry, is described. Recovery of added oestrogens was ~83%.

S. C. JOLLY.

Development of the digestive system of the young animal. III. Carbohydrase enzyme development in the young lamb. IV. Proteolytic enzyme development in the young lamb. D. M. Walker (*J. agric. Sci.*, 1959, 53, 374—380, 381—386).—III. Changes in amylase, lactase and maltase activity with advancing age in sheep are recorded. Utilisation of all carbohydrates except lactose and glucose by young lambs depends on the early development of the rumen flora and fauna.

IV. In the digestive organs of lambs, 1—5 weeks old, enzyme activity at pH 1.8 occurred only in the abomasum. All tissues were active on haemoglobin at pH 3.5. Activity on azocasein at pH 8.5 was found only in the contents of the pancreas and small intestines. In most tissues proteolytic activity increased with age. In the abomasum wall there was peak activity after 21 days with subsequent decline at pH 1.8 or 3.5.

A. G. POLLARD.

Crude protein requirements of the growing ewe lamb. R. W. Griffith, T. B. Keith and W. P. Lehrer, jun. (*Idaho agric. Exp. Sta.*, 1959, Res. Bull. 43, 15 pp.).—Lambs receiving diets containing 12.7–16.0% of crude protein made better gains and showed higher feed efficiency than did those receiving diets with 10.9% crude protein. Addition of 0.2% of DL-methionine to the 10.9%-protein diet improved wt. gains and feed efficiency, whilst addition of chlortetracycline (0.01 g. per lb. of feed) in addition increased wt. gains further, but reduced feed efficiency. A. H. CORNFIELD.

Range ewe production as affected by winter feed treatments. J. L. van Horn, O. O. Thomas, J. Drummond, A. S. Hoverland and F. S. Willson (*Montana agric. Exp. Sta.*, 1959, Bull. 548, 31 pp.).—Range ewes gained or lost wt. in direct proportion to the amount of supplemental feed supplied. Increasing feed level reduced the % of dry ewes and of lambs lost from birth to weaning, and increased body wt. gains during the winter, birth wt. of lambs, lb. of lamb weaned per ewe, fleece grease wt. and clean fleece wt. A. H. CORNFIELD.

Dehydrated lucerne in sheep production. J. L. van Horn, O. O. Thomas, J. Drummond, A. S. Hoverland, A. E. Flower and F. S. Willson (*Montana agric. Exp. Sta.*, 1959, Bull. 546, 18 pp.).—When ewes were wintered on average quality mixed hay a supplement (0.33 lb. of dehydrated lucerne hay or 0.5 lb. of 20%-protein pellets per head per day) for only 30 days before lambing was as satisfactory as feeding the supplement for the entire gestation period. The dehydrated lucerne hay supplement was more profitable than the pellets (soya-bean oil-meal + grain). A. H. CORNFIELD.

Protein supplementation of range sheep. J. L. van Horn, G. F. Payne, F. S. Willson, J. Drummond, O. O. Thomas and F. A. Branson (*Montana agric. Exp. Sta.*, 1959, Bull. 547, 15 pp.).—Body wt. of range-wintered ewes and % of live lambs increased with % protein in their pelleted supplementary feed. Although average birth wt. of lambs was slightly higher from ewes receiving the highest protein supplement, there were no differences in wt. at weaning. Wt. of lamb weaned per ewe increased with % of protein fed to the ewe. Clean fleece wt. tended to be higher when extra protein was supplied. A. H. CORNFIELD.

Digestibility of blue grama hay and production of fatty acids by sheep. K. A. Boller, M. W. Dross, W. W. Repp and W. E. Watkins (*New Mex. agric. Exp. Sta.*, 1959, Bull. 436, 13 pp.).—Addition of maize starch to the diet of sheep increased the digestibility of dry matter of blue grama. Ether extract digestion was reduced by addition of brewer's yeast, starch and CaHPO_4 and increased by addition of synthetic lucerne ash. Addition of urea or a "supplement" (maize starch + brewer's yeast + CaHPO_4 + synthetic lucerne ash) increased digestion of the N-free extract. Addition of urea + "supplement" increased digestibility of crude fibre and dry matter. Volatile fatty acid production was increased by addition of the "supplement." There was a close correlation between digestibility of crude fibre and dry matter and production of volatile fatty acids. A. H. CORNFIELD.

Determination of milk yield in merino ewe. I. McCance (*Aust. J. agric. Sci.*, 1959, 10, 839–853).—Lambs could obtain little or no milk from ewes hand-milked after two doses of 5 i.u. of posterior pituitary extract. Yields of milk depended on the interval between milkings, the rate of secretion being apparently faster in the first 2 hr. This effect was less marked in the later stages of lactation. Yields were independent of time of day and speed of milking. The order of yields among ewes was always significantly concordant. Earlier applications of the method had no effect on later lactation. The method provides a simple means of obtaining useful estimates of milk production. (17 references.) M. D. ANDERSON.

Effect of thyroxine implantation on wool growth. N. W. Godfrey and D. E. Tribe (*J. agric. Sci.*, 1959, 53, 369–373).—Implantation of thyroxine (90 mg.) in sheep receiving a restricted hay diet increased wool yields (12%) without affecting fibre diameter. No increase was produced in animals on an *ad lib* hay diet. Live wt. of all treated animals declined (10%) for about 7 weeks but after 10 weeks there was a gradual return to the initial wt. A. G. POLLARD.

Effect of supplementation with urea and molasses on the liveweight, appetite and wool growth of sheep. J. B. Coombe (*J. Aust. Inst. agric. Sci.*, 1959, 25, 299–301).—Addition of molasses and urea to a basal ration of ground oat straw increased the intake of straw by sheep, maintained the live-wt. of the animals (compared with 2 lb. per week loss in those receiving straw only) and somewhat increased wool yields. Supplements of molasses alone had no effect and larger additions of urea (>15 g. per head daily) caused no further response. A. G. POLLARD.

Present problems of pig breeding. Nutrition and infection. J. Blain and —. Joubert (*C. R. Acad. Agric., Fr.*, 1959, 45, 739–

740).—A review covering interactions between the incidence of major and (possibly latent) minor diseases, digestive disturbances affecting gains in wt., and the effects (especially on catabolism) of highly selective breeding. Increased attention to disinfection, feeding and immunisation is urged. P. S. ARUP.

Yield and composition of milk from sows fed [A] varying proportions of separated milk and concentrates : [B] on three ration levels. D. M. Smith (*N.Z. J. agric. Res.*, 1959, 2, 1057–1070; 1071–1083).—[A] Rations consisting of skim milk, with and without concentrates, were equally efficient in maintaining milk production by sows in N.Z., and both rations were more efficient than were concentrates alone. Milk energy production was more efficient in spring than in summer, while the efficiency of feed conversion by litters was higher in summer. Temp. conducive to efficient litter growth were antagonistic to efficient milk production. (15 references.)

[B] Reduction of the daily ration normal in N.Z. for lactating sows from 6 feed units (1 unit = 1 gal. of skim milk or 1 lb. of concentrates) plus 1 feed unit for each piglet suckled, to (4 + 2/3) units lowered milk yield slightly; a further reduction to (3 + 1/2) units caused no greater reduction. The reduction in yield was largely offset by increases in the energy content of the milk. Fat content of the milk tended to increase and lactose content decreased as the ration was reduced. Sows largely compensated for the smaller rations by using body reserves, and they used their food energy equiv. more efficiently on the lower rations. (17 references.) S. C. JOLLY.

Influence of feeding different levels of protein to porkers. P. J. S. Pieterse and W. A. Verbeek (*S. Afr. J. agric. Sci.*, 1959, 2, 343–355).—A maize-lucerne-wheat bran ration for pigs was supplemented with white fish meal in proportions up to 12.5%. Rates of gain in wt. and feed efficiency increased with the amount of fish meal used. With <15% of crude protein in the ration muscle development was restricted and deposition of fat increased. A. G. POLLARD.

Nutrition of goats. W. Hossain (*Agric. Pakistan*, 1959, 10, 299–307).—Maintenance requirements are determined in trials using sisal leaves and rape cake, detailed analyses and digestibility data for which are presented. A. G. POLLARD.

Practical ration for baby chicks. A. G. Hogan and R. W. Craghead (*Missouri agric. Exp. Sta.*, 1959, Res. Bull. 691, 19 pp.).—Chicks made satisfactory growth on diets in which maize and soya-bean oil-meal made up 96% of the total wt. and which contained added minerals and vitamins A, D and B_{12} and riboflavin. Addition of other vitamins, methionine, and other vegetable and animal proteins did not improve chick performance. A. H. CORNFIELD.

Poultry nutrition. W. Bolton (*Min. of Agric. Fish. Fd.*, 1959, Bull. 174, 90 pp.).—A review covering the nutrients required by poultry and the processes of digestion and absorption by which they are made available to the bird, energy metabolism and a new method of assessing the energy value of the diet, the quality of the proteins and the balances of vitamins and minerals in the diet, various feeding stuffs, use of antioxidants, antibiotics, etc., the formulation and the efficiency of utilisation of diets, and the diets used at various centres. H. S. R.

Chlortetracycline and arsanilic acid in rations for growing chicks. B. H. Schneider, I. A. Khan, M. Saleem and M. S. Shafiq (*Agric. Pakistan*, 1959, 10, 291–298).—Chlortetracycline (5 and 10 g./ton of feed) stimulated chick growth in only one of three trials. Arsanilic acid (0.01% of the ration) had no significant action. A. G. POLLARD.

Biological evaluation of the factors affecting the protein quality of fish meals. N. T. Rand, V. K. Collins, D. S. Varner and J. D. Mosser (*Poultry Sci.*, 1960, 39, 45–53).—An assay method based on chick growth and using "standard" protein and fish meal samples was used to evaluate the protein quality of commercial fish meal samples. Protein quality varied considerably and depended on the species of fish, type of organ, effect of heating, fat removal and duration of storage. A. H. CORNFIELD.

Influence of geographical area of production on chick growth response to barley samples subjected to enzyme supplements or water treatment. H. E. Willingham, K. C. Leong, L. S. Jensen and J. McGinnis (*Poultry Sci.*, 1960, 39, 103–108).—Chick growth was increased when diets containing barley grown in the West (U.S.) were supplemented with enzymes, but no increased growth occurred when barley grown in the Midwest and East was so supplemented. Chick growth was also increased by treating all samples of barley with water, the greatest response occurring with the Western barley. Responses of the barley samples to enzyme supplementation were not related to their proximate analysis. Eastern barley contained a heat-labile factor, probably enzymic. A. H. CORNFIELD.

Passage of weed seeds through the digestive tract of the chicken. J. B. Cooper, T. L. Maxwell, jun., and A. D. Owens (*Poultry Sci.*, 1960, **39**, 161—163).—Seeds of 25 varieties of weeds fed to chickens could not be detected in the faecal matter, nor was there any germination of weed seeds from faecal matter. This indicates that the chicken is not responsible for the spreading of weeds.

A. H. CORNFIELD.

Influence of fast and slow rises in ambient temperature on production traits and mortality of laying pullets. A. C. Campos, F. H. Wilcox and C. S. Shaffner (*Poultry Sci.*, 1960, **39**, 119—129).—Chickens subjected to an air temp. of 36.6° for 24 hr. showed a sudden temporary drop in egg production in some but not all breeds. There were no differences due to rate of temp. rise. Egg wt. and shell thickness decreased, these effects being more pronounced with the more gradual increase in temp. Albumin quality increased to a greater extent with the fast than with the slow rise in temp. Feed consumption decreased markedly, this effect being slightly greater with the faster rise in temp. The treatments had no consistent effects on mortality.

A. H. CORNFIELD.

Amino-acid requirement of the growing chick fed a crystalline amino-acid diet. G. J. Klain, H. M. Scott and B. C. Johnson (*Poultry Sci.*, 1960, **39**, 39—44).—A chick diet based on crystalline amino-acids supported much faster growth than did other reported diets. The requirements for chick growth of 12 amino-acids are presented.

A. H. CORNFIELD.

Amino-acid requirement of laying hens. V. Amino-acid balance in low-protein diets. H. Fisher, P. Griminger and H. Lutz (*Poultry Sci.*, 1960, **39**, 173—175).—Laying hens on a 12% protein diet low in arginine showed improved body wt., egg production and egg size when the diet was supplemented with 2.5% of gelatin (source of arginine), whilst 2.5% of glutamic acid depressed body wt. and egg production. In a chick growth study using the same diets growth was improved by gelatin and depressed by glutamic acid.

A. H. CORNFIELD.

Changes in plasma iron, haemoglobin and plasma proteins in immature pullets resulting from simultaneous administration of (a) oestrogen and thyroxine and (b) oestrogen and sulphamethazine. E. A. Campbell (*Poultry Sci.*, 1960, **39**, 140—144).—Thyroxine (0.001 g. per bird per day) inhibited the changes in haemoglobin and serum-Fe produced by oestrogen (oestradiol benzoate, 0.018 g. over 14 days) whilst sulphamethazine (0.2% in the feed) augmented the effect of the oestrogen. The rise in serum-protein following oestrogen administration was accompanied by a lowering of serum-albumin and increases in the β - and γ -globulin fractions. These effects were inhibited by thyroxine and augmented by sulphamethazine.

A. H. CORNFIELD.

Bioassay for determining the nutritional adequacy of protein supplements for chick growth. S. W. Hinners and H. M. Scott (*Poultry Sci.*, 1960, **39**, 176—183).—The bioassay method, based on chick growth, is described and is shown to be satisfactory in demonstrating the nutritional value of various fish meals.

A. H. CORNFIELD.

Barley in high-efficiency broiler rations. IV. Influence of amylolytic enzymes on efficiency of utilisation, water consumption and litter condition. G. H. Arscott and R. J. Rose (*Poultry Sci.*, 1960, **39**, 93—95).—Growth of broiler chicks to 8 weeks of age was somewhat lower when the maize in a 61% grain mash ration was replaced with barley. When the barley mash diet was pelleted or treated with amylolytic enzymes growth was much better, but still not comparable with that on the maize mash. Growth comparable to that with maize mash was obtained when the barley mash was treated with both enzymes and animal fat or was pelleted after addition of enzymes. Water consumption was increased with all the diets containing barley in comparison with the maize mash diet. Litter condition was excellent for the maize mash, good for the barley + enzyme diets, and fair to poor for the other barley diets.

A. H. CORNFIELD.

Effect of dehydrated lucerne meal, dried brewer's yeast, condensed fish solubles and fermentation residue on the reproductive performance of turkeys. C. H. Whiteside, T. M. Ferguson, B. L. Reid and J. R. Couch (*Poultry Sci.*, 1960, **39**, 77—81).—Addition of 5% of lucerne meal, dried brewer's yeast or condensed fish solubles, or 1% of fermentation residue (containing streptomycin sulphate 1.5 and vitamin B₁₂ 0.0005 g. per lb.) to the diet (all-vegetable) of turkey hens increased hatchability by 15.1—21.8% and average egg wt. by 2—7 g. Dried brewer's yeast was the only supplement which increased egg production and feed efficiency with respect to egg production.

A. H. CORNFIELD.

Relationship between fat, unidentified factors and pelleting of diets for chickens and turkeys. W. F. Pepper, S. J. Slinger and J. D. Summers (*Poultry Sci.*, 1960, **39**, 66—74).—Addition of 2.5—5.0% of fat to diets in mash form increased wt. gains of birds, but did not

do so when added to similar diets in pelleted form. Pelleting usually increased wt. gains, the increase being greatest in the absence of added fat and declining with level of added fat. Both pelleting and addition of fat improved feed efficiency. There is a sparing relationship between fat and pelleting and also probably between fat and unidentified growth factors.

A. H. CORNFIELD.

Influence of body weight on the experimental production of perosis by manganese deficiency. R. D. Creek, H. E. Parker, S. M. Hauge, F. N. Andrews and C. W. Carrick (*Poultry Sci.*, 1960, **39**, 96—98).—Hock tendons were severed in the right limbs of chicks at 3 days of age. There was high incidence of perosis in the left leg even when dietary Mn was apparently adequate (100 p.p.m.). The degree of perosis on Mn-deficient diets (7—20 p.p.m.) was much less severe in the immobilised leg than in the legs of normal chicks.

A. H. CORNFIELD.

Effect of technical grade manganese sulphate on vitamin stability in stored feeds. R. D. Creek, C. W. Carrick, S. M. Hauge and H. E. Parker (*Poultry Sci.*, 1960, **39**, 109—111).—Addition of Mn (200 p.p.m. technical MnSO₄ to a complete feed or 746 p.p.m. to a concentrate) did not affect the stability of thiamine, choline, carotene, niacin, Ca pantothenate or riboflavin during storage for 4 months.

A. H. CORNFIELD.

Vitamin-E activity of selenium in turkey hatchability. C. R. Creger, R. H. Mitchell, R. L. Atkinson, T. M. Ferguson, B. L. Reid and J. R. Couch (*Poultry Sci.*, 1960, **39**, 59—63).—Turkey hens were fed a vitamin-E-deficient diet for the first 9 weeks of the laying period. Subsequent supplementation of the diet with vitamin E (10 i.u. per lb. of feed) increased fertility and hatchability of fertile eggs. Dried brewers' yeast (5%) increased fertility but not hatchability, whilst Se (0.1 p.p.m.) depressed fertility slightly and had no effect on hatchability.

A. H. CORNFIELD.

Conversion of β -carotene to vitamin A in vivo. I. Time required for conversion to take place. II. Existence of an intermediary product(s). I. R. Sibbald and L. M. Hutcheson (*Poultry Sci.*, 1960, **39**, 99—103).—When β -carotene was injected into the ligatured duodenal loops of vitamin-A-deficient chicks, some birds produced vitamin A in less than 5 min., whilst the majority converted carotene to vitamin A within 15 min. The relationship between the carotene conversion ratio and amount of vitamin A produced indicates the presence of at least one intermediary compound in the conversion of carotene to vitamin A. The min. quantity of β -carotene required to produce 1 i.u. of vitamin A was 0.383 \pm 0.685 μ g.

A. H. CORNFIELD.

Influence of dietary energy level, energy source and breed on the thiamine requirement of chicks. P. A. Thornton and J. V. Shutze (*Poultry Sci.*, 1960, **39**, 192—199).—Both light and heavy breeds of chickens made normal growth with 0.0004 g. of thiamine hydrochloride per lb. of diet. Increasing the energy content of the non-supplemented diet by addition of sucrose intensified thiamine deficiency to a greater extent than did addition of maize oil. The light breed had a somewhat higher thiamine requirement than had the heavy breed in that mortality was higher and survival time was shorter with the former on non-supplemented diets.

A. H. CORNFIELD.

Effect of high levels of dietary cholesterol on the serum proteins of the chicken. R. E. Clegg, A. T. Ericson and U. K. Misra (*Poultry Sci.*, 1960, **39**, 35—39).—Electrophoretic patterns of the blood serum proteins of cholesterol-fed birds showed distinct differences from proteins of birds given injections of diethylstilboestrol. In particular cholesterol feeding increased the amounts of serum-electrophoretic components 4 and 5 and the P content.

A. H. CORNFIELD.

Effect of antibiotic supplementation on the response of poult to dietary maize oil. W. C. Supple (*Poultry Sci.*, 1960, **39**, 227—229).—Increasing the maize oil level of the diet from 4% to 13.3% increased poult growth to 3 weeks of age by 20% in the presence of oleandomycin phosphate (0.05 g. per kg. of feed) and by 10% in the absence of the antibiotic. Addition of 18.7% of maize oil to the diets resulted in somewhat reduced wt. gains, in comparison with the 13.3% level, both in the absence and presence of antibiotic. The response to antibiotic increased from 14% with the 4% maize oil diet to 27% with the 18.7% maize oil diet.

A. H. CORNFIELD.

Interaction between breed and feeding treatment for egg production. S. Fox, R. C. Jennings, A. Marsden and T. R. Morris (*Poultry Sci.*, 1960, **39**, 235—236).—Egg production by Rhode Island Red \times Light Sussex pullets was higher with a mash diet (910 kg.-cal. per lb., 16.4% crude protein) than with a pelleted diet (1010 kg. cal. per lb., 18.0% crude protein), whilst the reverse held for a type of small hybrid (similar in appearance to White Leghorns).

A. H. CORNFIELD.

Relationship of rate of egg production as affected by feed to Haugh units of eggs. R. H. Harms and C. R. Douglas (*Poultry Sci.*, 1960,

39, 75—76).—There was a significant negative correlation between rate of egg production and Haugh units (albumin quality) of eggs from hens receiving varying dietary protein levels. Increased egg production arising from feeding antibiotics was accompanied by decreased Haugh units in the eggs. A similar relationship occurred when the P content of the hens' diet was varied.

A. H. CORNFIELD.

Control of infectious synovitis. VII. Comparison of tetracycline antibiotics. D. C. Shelton and N. O. Olson (*Poultry Sci.*, 1959, 38, 1309—1315).—When given intraperitoneally 0.001 g. of either chlortetracycline or oxytetracycline per 100 g. body wt. gave effective control of infectious synovitis in chicks, whereas orally 0.0012 g. of chlortetracycline and 0.0022 g. of oxytetracycline per 100 g. body wt. was required. Tetracycline requirements were somewhat higher (exact amounts not determinable from data) both intraperitoneally and orally, with the latter method of administration being only 40% as effective as the former. A. H. CORNFIELD.

Depletion of insecticides in sheep dipping baths. I. R. Harrison and P. G. Marshall (*J. Sci. Fd Agric.*, 1959, 10, 568—576).—Factors affecting the depletion rates of, e.g., aldrin, dieldrin and γ -BHC from sheep dipping baths under field conditions are discussed. Where the initial concn. is high, rapid depletion occurs. Theoretically it seems better to start at a lower concn. and increase the amount added at topping up to give a constant overall concn. Increased depletion occurs when lambs with a heavier fleece are dipped compared with sheep carrying little wool. Thorough mixing of the bath before dipping is very important. Details are given of sampling and i.r. analysis of the insecticide. E. M. J.

Chemical and physiological investigation of geeldikkop in sheep in South Africa. J. M. Brown and W. T. de Kock (*S. Afr. industr. Chem.*, 1959, 13, 189—191).—The extraction and characterisation of the saponins present in *Tribulus terrestris* L. are described. This plant is believed to be the cause of geeldikkop in sheep, but when sheep were dosed with the unhydrolysed and partially hydrolysed saponins all the typical symptoms of the disease were not observed. (16 references.) A. M. SPRATT.

Korlan for sheep scab control. J. W. Trumble and R. G. Howe (*Down to Earth*, 1959, 15, No. 1, 14—15).—When used as a single sheep-dip Korlan-25W (a wettable powder containing 25% of *OO*-dimethyl *O*-2,4,5-trichlorophenyl phosphorothioate) in concn. 0.5% of active ingredient effectively controlled scab and stimulated new wool growth. A. G. POLLARD.

In-vivo testing of chemicals for control of cattle grubs. P. H. Kohler (*Dissect. Abstr.* 1959 20, 1935).—Using 11 methods of application to cattle, 14 substances were tested for control of the cattle grubs *Hypoderma bovis* and *H. lineatum*. Ronnel by mouth, and Co-Ral and Dowco 109 by external application, were effective, and did not affect gains of wt. in calves, or cause toxic symptoms. Dimethoate was effective by mouth or intramuscularly, but caused some toxic symptoms, and delayed gain of wt. Nicarbazine at effective doses was toxic. Phenothiazine and Hypolin, given by mouth, were ineffective. M. D. ANDERSON.

Ruelene for cattle grub control: progress report. W. S. McGregor, P. D. Ludwig and L. L. Wade (*Down to Earth*, 1959, 15, No. 2, 2—3).—Use of Ruelene (4-t-butyl-2-chlorophenyl methyl methylphosphoramidate) in the diet of cattle and sheep, (7.5 mg./kg. body wt./day), or as a spray (0.75% applied at 150 p.s.i.) gave excellent control of hornworm, screw worm, lice, etc. Multiple doses, given orally, were superior to single large doses. A. G. POLLARD.

Weight gains and grub control in cattle treated with Trolene. M. G. Norris and R. G. Howe (*Down to Earth*, 1959, 15, No. 2, 4—5).—Trolene (a purified grade of *OO*-dimethyl-2,4,5-trichlorophenyl phosphorothioate) at the rate of 5 g./100 lb. live wt. given as a bolus or mixed with the feed gave 90% control of cattle grubs and, in some cases also, of lice. A. G. POLLARD.

DDT, lindane and malathion for control of long-nosed cattle louse, *Ligognathus vituli* (L.). D. W. Anthony (*J. econ. Ent.*, 1959, 52, 782).—Although a great reduction in the residual effectiveness of DDT and lindane but not of malathion on calves was noted, laboratory test on the lice failed to show resistance. C. M. HARDWICK.

Control of mange mites in laboratory white mice with malathion. J. E. Webb, jun., and E. L. Shepherd (*J. econ. Ent.*, 1959, 52, 790).—Application of 25% malathion dust, once a week for 6 weeks, to the bedding of the mice eradicated *Myocoptes musculinus*. Malathion emulsion was also satisfactory. The use of 75% of lindane powder killed the mice. C. M. HARDWICK.

Dowco 109 as an animal systemic insecticide. R. O. Drummond and O. H. Graham (*J. econ. Ent.*, 1959, 52, 749—750).—Screening tests showed that subcutaneous injection of Dowco 109 [*O*-(4-t-butyl-

2-chlorophenyl) *O*-methyl methylphosphoramidothioate] killed screwworm larvae in guinea pigs. A drench of 50 mg./kg. killed screwworm larvae in sheep. Drenches of 15 mg./kg., and injection of 25 mg./kg. reduced numbers of first-instar *Hypoderma* spp. in the backs of cattle by 90%. C. M. HARDWICK.

Rotenone and methoxychlor dust in backrubbers for horn fly control in dairy herds. L. T. Hargett and R. L. Goulding (*J. econ. Ent.*, 1959, 52, 762—763).—Backrubbers charged with 5% rotenone gave 6—7 weeks' satisfactory control, those containing 5% methoxychlor 3—5 weeks. Malathion dust (4%) and 25% Delphene (*NN*-diethyl-*m*-toluamide) dust lasted only one week. The average doses per cow were 0.9 oz. of rotenone every 30 days and 0.03 oz. of methoxychlor every 21 days, this being less than current recommendations. C. M. HARDWICK.

Ronnel and Co-Ral for horn fly control on cable-type backrubbers. E. C. Burns, S. E. McCraime and D. W. Moody (*J. econ. Ent.*, 1959, 52, 648—650).—Backrubbers, treated once in 2 weeks with 5% toxaphene, 0.25% Co-Ral [*O*-(3-chloro-4-methylumbelliferone) *OO*-diethyl phosphorothioate] or 1% Ronnel (Dow ET-57), kept horn fly populations at a low level. C. M. HARDWICK.

Control of fowl cholera. T. A. Dorsay and G. S. Harshfield (*S. Dakota agric. Exp. Sta.*, 1959, Tech. Bull. 23, 18 pp.).—Addition of oxytetracycline (0.5—1.0 g. per kg. of feed) to the diet of infected hens was as effective as was 0.5% of sulphamerazine or 0.05% of sulphaquinoxaline in the feed in reducing mortality due to fowl cholera. Penicillin and streptomycin (1 g. per kg. of feed) were ineffective. A. H. CORNFIELD.

Malathion dust for control of lice on domestic game birds. J. L. Rodriguez and L. A. Riehl (*J. econ. Ent.*, 1959, 52, 774—775).—Malathion (1%) in the dusting wallows killed all lice within 7 days and kept the birds free from lice for >90 days. C. M. HARDWICK.

Malathion dust for control of two species of pigeon lice. J. L. Rodriguez and L. A. Riehl (*J. econ. Ent.*, 1959, 52, 772).—Dusting pigeons with 0.5 or 1% malathion or putting 1% malathion in the nests gave >4 months' control of *Lipurus baculus* and *Gonicocotes bidentatus*. C. M. HARDWICK.

Isolation and cultivation of some fungi from soils and pastures associated with facial eczema disease of sheep. R. H. Thornton and D. J. Ross (*N.Z. J. agric. Res.*, 1959, 2, 1002—1016).—The species of fungi isolated from areas in N.Z. with a known history of facial eczema of sheep are reported. Some cultures of *Sporidesmium bakeri* produced symptoms in the liver associated with facial eczema when fed to guinea-pigs. (26 references.) S. C. JOLLY.

Controlling infectious bronchitis in Maine chickens. H. L. Chute, D. C. O'Meara and J. F. Witter (*Maine agric. Exp. Sta.*, 1959, Bull. 584, 26 pp.).—Results obtained with a vaccination programme of 6,000,000 birds over 8 years are presented. A. H. CORNFIELD.

Effects of administration of thiocyanate and thiouracil to sheep. E. Wright (*N.Z. J. agric. Res.*, 1959, 2, 903—914).—The unpredictable toxicity of CNS⁻ to sheep appeared to be due to failure of urinary CNS⁻ excretion, with resulting development of lethal blood-CNS⁻ levels. Smaller goitres were produced in lambs by the administration of high levels of methylthiouracil than by intermediate levels; at the higher levels, I⁻ enhanced goitrogenesis, while at the lower levels it was partially prophylactic. Duration and time of goitrogen administration affected ¹³⁷I⁻-concn. test. Previous suggestions that carrier I⁻ could increase concn. of ¹³⁷I⁻ in sheep by overcoming the goitrogenic effect of low levels of dietary CNS⁻ were not confirmed. Regular ingestion of thiouracil by pregnant sheep interfered with the rapid development of foetal thyroid activity that occurs in the fourth month of pregnancy. S. C. JOLLY.

Animal feeds. C. Pfizer & Co. Ltd. (B.P. 812,416, 15.12.55, U.S. 15.12.54 and 3.2.55).—A growth-accelerating, animal feed supplement comprises a tetracycline-type or penicillin antibiotic (50—98) and an orally active oestrogen (2—50 wt. %). The composition may also contain protein (10—14), fat (2.5—3), fibre (16—10), minerals (5—6), vitamins and nitrogen-free extract (65—68%) suitable for cattle and other ruminants. Thus, incorporation of oxytetracycline (10) and diethylstilboestrol (1 g.) into cattle feed (1 ton) results in an average daily gain in wt. of 0.4 as compared with only 0.31 lb. for control animals. F. R. BASFORD.

Animal feed supplements for treatment of gastro-intestinal parasitic diseases. Dow Chemical Co. (B.P. 802,512, 2.12.55, U.S., 10.12.54 and 22.8.55).—A concentrate for incorporating into animal feed comprises 5—95 wt.-% of a dinitrophenylurea compound NRR'CO-NXH (R and R' are H or alkyl of 1—4 C; X is 2,4-

3,5-dinitrophenyl), a non-toxic carrier (solid or oil), and optionally essential minerals, vitamins, etc. F. R. BASFORD.

Growth regulating composition. K. H. Jaeger and H. Mittenzwei (B.P. 809,505, 24.7.55. Ger., 27.4.54).—One or more than one organ of a living (vertebrate warm-blooded) organism (e.g., the thymus, spleen, etc.) is subjected to artificial stimulation (exposure to radiations of various types; treatment with chemicals and radioactive Co, I or P), to induce in the parenchyma or mesenchyma thereof a state of unusual activity, then the organ is extracted to recover a protein-free growth regulant prep. consisting of nucleic acid derivatives, amino-acids and polypeptides.

F. R. BASFORD.

Parasitical drugs. National Research Development Corp. (Inventor: J. Williamson) (B.P. 809,295, 17.3. and 27.7.56).—Compounds useful in the treatment of trypanosomiasis in animals (cattle, pigs, horses) comprise salt complexes (of low water-solubility) of Suramin and a basic trypanocidal drug, viz., Antrycide dimethyl sulphate, Ethidium bromide, a salt of 2-amino-10-methyl-7-(2'-amino-1,6'-dimethylpyrimid-4'-ylamino)-9-*p*-aminophenylphenanthridine [e.g., the dibromide (= RD. 2801)], or a salt of 2-amino-10-ethyl-7-(2'-amino-1,6'-dimethylpyrimid-4'-ylamino)-9-*p*-aminophenylphenanthridine [e.g., the dimethanesulphonate (= RD. 2902)]

F. R. BASFORD.

Hormone preparations. CIBA Ltd. (B.P. 809,827, 23.6.55. Switz., 2.7.54).—A hormone prep. containing testosterone undecylate (15), valerate (8) and propionate (2 pt.) is claimed. The prep. may be employed in the form of a tablet or an ampouled solution containing 0.5—500 mg. of mixture for use in veterinary medicine.

F. R. BASFORD.

Control of parasites in animals. Dow Chemical Co. (B.P. 809,814 8.2.57. U.S., 28.3.56).—A composition, for use in the control of parasites (especially cattle grub) in warm-blooded animals comprises nutrient feed containing 0.01—95 wt.-% of a compound OR(OR')·PS·X or (OR)₂PS·OR'' (R is Me or Et; X is NH₂, NHMe or NHEt; R' is aryl, viz., *p*-methoxyphenyl, *o*-chloro-*p*-alkylphenyl or halogenophenyl; R'' is halogenophenyl, viz., monochloro-, 2,4-, 2,5- or 3,4-dichloro-, or 2,4,5-trichlorophenyl). One example is *OO*-dimethyl *O*-2,4,5-trichlorophenyl phosphorothionate.

F. R. BASFORD.

Substantially anhydrous ferrous fumarate. Mallinckrodt Chemical Works (B.P. 807,638, 14.3.57. U.S., 18.5.56).—Anhydrous ferrous fumarate (for use as haematinic in animal feed supplement) is obtained by admixing a water-sol. Fe^{II} salt with a water-sol. fumarate in aq. medium at >70°.

F. R. BASFORD.

New veterinary compositions. Imperial Chemical Industries Ltd. (Inventor: W. Hepworth) (B.P. 806,725, 2.5.56).—A composition, useful in the treatment of coccidiosis in fowl, contains as active ingredient 0.005—0.5 wt.-% of a compound *p*-NO₂C₆H₄·NH·CO·NH·C₆H₄X·*p*' (X is halogen other than Br). The composition may be in the form of a powder (suitable for incorporating into feeding stuffs), a dispersible powder (containing wetting agent) or an aq. dispersion (containing additional non-toxic excipients, e.g., sweetening agent such as lactose, and solubilising agent such as propylene glycol). The composition may also be made up in the form of capsules or tablets. Thus, 4-chloro-4-nitrocarbanalide is admixed (2) with chicken mash (10,000 pt.), to provide a composition suitable for use as sole ration for chicks—to render them immune to infection by *Eimeria tenella*.

F. R. BASFORD.

5,5'-Dinitro-2,2'-dichlorobenzil. Société des Usines Chimiques Rhône-Poulenc (Inventors: H. Moreu and P. Chovin) (B.P. 807,985, 7.6.57).—The compound, prepared by nitration of 2,2'-dichlorobenzil, is claimed as being useful in veterinary medicine, e.g., against coccidiosis.

J.A.C. ABSTR.

Pharmaceutical and veterinary preparations containing garlic. H. Schiefer (B.P. 808,568, 21.11.55. Aus., 4.11.54).—A medicinal prep. for use in the blood treatment of human beings and animals, is prepared by extracting finely comminuted garlic during 24—36 hr. at or above room temp. with 40—50 vol.-% aq. EtOH, then adding (to the filtered extract) lemon juice and anethole, refiltering after 8—18 hr., then further adding comminuted bananas, honey and pressed yeast.

F. R. BASFORD.

2.—FOODS

Carbohydrate Materials

Cereals, flours, starches, baking

Indian rice varieties. P. F. Pelshenke and G. Hampel (*Getreide u. Mehl*, 1960, 10, 29—32).—Biometric, chemical and physical ex-

aminations were carried out on 26 varieties of Indian rice, husked and polished in the laboratory. Samples of only two varieties, T 100 and Patnai 23, were of commercial grain size. The 1000-grain wt. and vol. afforded reliable measurements of size. Ash varied 0.32—0.63%, protein 5.8—10.3%, sol. carbohydrates 0.8—2.6%. The members of each of two groups (red-husked varieties and the three smallest varieties) showed values for sol. carbohydrate, swelling value and swelling vol. which suggest a rather close systematic association. Viscogram and cooking results ranged rather widely. There was some indication of a relation between cohesion of the cooked rice grains and the η min. at 90° of the viscogram, confirming an observation already made with commercial polished white rice. (10 references.) C. L. HINTON.

Use of emulsifiers and emulsified oils to reduce cohesion in canned white rice. R. E. Ferrel, E. B. Kester and J. W. Pence (*Food Technol.*, 1960, 14, 102—105).—Treatment of rice with surface-active materials and vegetable oil emulsions during canning reduced cohesion between kernels without affecting other properties of canned rice which make it superior to freshly cooked rice. Cold storage after canning was beneficial to the product. Reduction of cohesion was directly related to the logarithm of oil concn. in the emulsions. No change in organoleptic properties appeared in treated rice after 19 weeks of holding at 100°F. E. M. J.

Wheat sizing technique for predicting flour milling yield. W. C. Shuey (*Cereal Sci.*, 1960, 5, 71—72, 75).—Sizing of wheat according to its cross-sectional area gives a more reliable indication of milling yield than does the test wt. of lb. per bushel. The mechanical wheat sizer is described. Correlations are given between sizing yield, mill yield and test weight over a large number of samples.

S. G. AYERST.

Uniform quality appraisal of coarse and fine semolinas for the manufacture of alimentary pastes. B. Pagenstedt (*Getreide u. Mehl*, 1960, 10, 32—36).—The properties of semolinas which are significant in relation to their suitability for the manufacture of alimentary pastes are discussed. They include fineness and size distribution, acidity, fibre or speck content, yellow pigment or colour, content of protein and wet or dry gluten and its characteristics, lipoxidase activity, water absorption, dough consistency and swelling capacity, and preliminary determination of cooking properties. Moisture and ash content are of chief interest for control purposes. Methods of test at present available are indicated, and stress is placed on the desirability of improving and developing these objectively.

C. L. HINTON.

Diastase methods for wheat and rye flours. S. Hagberg (*Brot u. Gebäck*, 1960, 14, 41—47).—Methods available for the determination of diastatic condition in flour (embracing both amylase activity and starch character) are reviewed, in relation firstly to proving of dough and secondly to quality of crumb in the bread. Methods for separate determination of amylase activity and starch character are also discussed. A viscometric method is proposed for determining the pasting and liquefaction times of the starch, and values for normal and high activity flours are quoted. From the difference between the two values a figure for "diastase value" can be calculated. (27 references.) C. L. HINTON.

Promoters of sour dough fermentation. G. Spicher and H. Stephan (*Brot u. Gebäck*, 1960, 14, 47—52).—In laboratory and baking tests representative strains from eight groups of lactic acid bacteria (I), isolated in previous work from sour dough, were compared as to their influence in the souring of dough and their effects on the quality of the bread. The micro-organisms, after enrichment in sterile rye mash, were added to the sour dough mixture both direct and after preliminary refreshing with flour and water, as a starter. Under the conditions used, I of different groups showed different effects on the sour dough, particularly in the course and degree of souring and the quality of the bread subsequently prepared. With favourable operating conditions, sour doughs satisfactory for baking purposes could in all cases be obtained, although only heterofermentative bacteria gave outstanding doughs of typical aroma. The view that different kinds of I have characteristic properties of importance for their effects in sour dough is confirmed. (13 references.) C. L. HINTON.

Inhibitory effect of acetic acid and sodium benzoate on the growth of two strains of osmophilic yeast. M. S. S. Rao and D. S. Johar (*Food Sci., Mysore*, 1959, 8, 383—386).—Growth of the osmophilic yeasts, *Saccharomyces mellis* and *Saccharomyces rouxii*, in a syrup containing sucrose, inorg. salts, and yeast extract, was respectively inhibited by addition of 0.4 and 0.5% of acetic acid. In presence of 50 p.p.m. of Na benzoate, only 0.2 and 0.3% of acetic acid were required to inhibit the growth of inocula of 2 million cells per ml., and on transfer to glucose broth the cells were found to be non-viable. (12 references.) M. D. ANDERSON.

Chromatographic fractionation of polysaccharides on cellulose anion-exchange columns. H. Neukom, H. Deuel, W. J. Heri and W. Kündig (*Helv. chim. Acta*, 1960, **43**, 64—71).—Polysaccharides are adsorbed on diethylamino-cellulose from solutions of different pH according to their content of acid groups. Neutral polysaccharides are only weakly adsorbed at pH 6 and are readily eluted at the same pH, but are adsorbed by the hydroxyl form of the cellulose and can be eluted fractionally at increasing pH. Acid polysaccharides are adsorbed at pH 7 and eluted $>$ pH 7; use of the borate form of the cellulose and borate buffers provides better fractionation. Analyses are presented of various pectic acids, dextrin, pentosans and albumins. A sugar-beet pectic acid is shown to contain arabin and galactan covalently linked to the polygalacturonic side chains. A polysaccharide-protein complex containing xylose, arabinose and galactose is present in wheat flour. J. L. PROSSER.

Role of lipids in baking. R. L. Glass (*Cereal Sci.*, 1960, **5**, 60—62, 75).—Flour lipids appear to be a vital factor in flour quality. Their constitution, interactions and rôle in the baking behaviour of flour is only partly known. The present state of knowledge is reviewed, concerning identification techniques, bound and extractable lipids, the lipoprotein complex, lipid binding in gluten, reactions during baking, effect on O_2 uptake in doughs, and the relationship of lipids to bromate action in doughs. (17 references.) S. G. AYERST.

Food products. G. Fioravanti (B.P. 813,731, 21.1.57. It., 19.1.56).—The deterioration of colour of wheat semolina dumplings ("gnocchi") in storage is obviated by preheating the semolina and dry starch to 30—70° before the dough is made. E. ENOS JONES.

Wet starch manufacturing process. Corn Products Refining Co. (B.P. 811,686, 14.3.57. U.S., 13.11.56).—A process for the manufacture of a starch product, in which the elimination of certain stages is made feasible, comprises subjecting starch-bearing grain (especially maize or sorghum grain) to steeping and cracking and germ removal to provide a wet degerminated material ($<$ 65 wt.-% of water) containing the hull and unmilled endosperm; feeding the material into a high-speed rotor disc rotating at a high speed (peripheral velocity $<$ 21,000 ft. per min.), to throw it on to a surface, thereby separating the gluten and starch from the fibre without shredding the latter. More specifically, the rotor is equipped with rotor pins about 0.75 in. in diameter and about 0.5—1.5 in. apart. F. R. BASFORD.

Starch phosphate. International Minerals & Chemical Corp. (Inventor: [A] H. Neukom (B.P. 812,339—40. [A] 15.8, [B] 25.8.55. [B] U.S., 16.9.54).—[A, B] Starch is heated with an aq. solution (pH 3—7.5) of at least one alkali metal orthophosphate (NaH_2PO_4) for a time sufficient to incorporate 1—2% of P into the starch mol. Unabsorbed fluid is removed and the residue heated for 1—15 hr. at 120—175°. [B] 2—5% of urea is added with the phosphate solution before heating. [A] The product has thin-binding properties. [B] The paste from the product has much shorter properties than that from the product from [A]. J. A. C. ABSTR.

Pudding mix containing alkali metal starch phosphate. International Minerals & Chemical Corp. (B.P. 812,363, 15.8.55. U.S., 16.9.54).—A pudding composition, adapted to produce a pudding almost instantaneously on admixture with cold milk comprises sugar, flavouring and a cold-water-swelling alkali metal starch phosphate. The latter, containing 1—5 wt.-% of bound P and 1—1.75 atoms of alkali metal per atom of P, is obtained by admixing ungelatinised starch with an aq. solution (pH 4—7) of an alkali metal orthophosphate, removing unabsorbed liquid from the treated starch below the temp. of gelatinisation, then heating at 120—175 (140—170°) until the reaction is complete (1—15 hr.). F. R. BASFORD.

Pudding compositions. Monkhouse & Glasscock Ltd. (Inventor: M. F. Alexander (B.P. 813,725, 28.6.56).—A pudding composition (which on addition to cold milk affords within 5—10 min. a product closely resembling a cooked starch pudding) comprises sugar (74), pregelatinised starch (19), flavouring (0.6%) and (as milk protein coagulant) 1—10% of a dialkali pyrophosphate, e.g., $Na_2H_2P_2O_7$ (3.2), and (as coagulation accelerator) 1—10% of a trialkali metal orthophosphate, e.g., Na_3PO_4 (3.2%). F. R. BASFORD.

Sweetening compositions. Abbott Laboratories (B.P. 812,575, 29.8.56. U.S., 29.8.55).—A sweetening composition (in the form of a tablet or aq. solution) comprises a cyclohexylsulphamate (preferably the Ca or Na salt) (15—50), a saccharin salt (Na or Ca salt) (5—15 mg.) and a carrier (e.g., Na benzoate). The composition is such that in aq. solution the concn. of the sulphamate and saccharin fall within defined limits. F. R. BASFORD.

Sugars and confectionery

Sugar distribution in plant tissues cooked in syrup. C. Sterling and C. O. Chichester (*Food Res.*, 1960, **25**, 157—160).—Precise cellular distribution of radioactive sugars was followed in canned clingstone peaches. Microscopical sections were compared with autoradiographs made from the sections. The cell walls showed the highest radioactivity and the vascular bundles had the highest concn. of radioactive sugar. Data indicate an adsorption of the sugars by the cell wall, probably against a concn. gradient. E. M. J.

Determination of reducing sugars by back-titration against alkaline copper solution. I. M. Takahashi (*Bull. chem. Soc. Japan*, 1960, **33**, 178—181).—A volumetric determination by titrating a boiling mixture of modified Fehling solution and sugar solution with a standard sugar solution is described. The method is applicable over a wide range of sugar concn., and especially to dil. solutions, with high accuracy. I. JONES.

Extraction of sugar from sugar cane. National Cylinder Gas Co. (B.P. 813,394, 10.1.56. U.S., 27.1.55).—A process for the extraction of sugar-bearing juice from sugar cane by continuous diffusion comprises cutting the cane into pieces whose dimension along the fibre axis is no greater than the transverse dimension of the cane stalk; suspending the pieces in sugar-bearing juice (1—2 vol. per vol. of pieces), preferably hot juice ($<$ 65°) of 10—17° Brix; introducing the resulting suspension near the bottom of a diffusion tower up which the cane pieces are moved countercurrent to a stream of hot water ($<$ 65°); removing exhausted cane at the top of the tower; recycling part of the collected sugar-bearing juice; and withdrawing the remaining juice (from the bottom of the tower). The pieces of sugar cane, which should traverse the tower during 20—45 min., are preferably circular in shape, 3—10 mm. in thickness, and $>$ 13 mm. in length. F. R. BASFORD.

Confection. National Dairy Products Corp. (Inventors: A. J. Doumak and J. Doumak) (B.P. 813,717, 6.5.57).—A method of making a confection, especially a marshmallow, comprises beating a mixture of starch, saccharides, gelatin and moisture (13—19%) under gas pressure (175 lb. per sq. in.) to provide a product of density 30—65 oz. per gal. The latter is then extruded into a zone of quiescence (at 105—130°r) and is cut prior to gelling. F. R. BASFORD.

Emulsions of phosphatides in aqueous alcohol. C. H. Buer Chemisch Pharmazeutische Fabrik (B.P. 813,213, 12.12.56. Ger., 17.12.55).—An emulsion of a phosphatide in aq. alcohol ($>$ 25% of alcohol) containing a carbohydrate (e.g., pentose or hexose, a disaccharide, or a polysaccharide) is stabilised by incorporation of a neutral fat (0.5—5 wt.-% on phosphatide). Thus, fat-free choline-calamine-lecithin (100) and oil (1) contained in lecithin are emulsified at 80° in a solution of dextrose in 15% aq. EtOH, to provide a completely stable emulsion (particle size of lecithin 0.03—0.06 μ), suitable for use in the chocolate and confectionery industries, in the prep. of cosmetic creams, and in the treatment of hides. An emulsion similarly formed in absence of oil is unstable. F. R. BASFORD.

Fermentation and Alcoholic Beverages

Effect of sulphur dioxide and fermentation on colour extraction from red grapes. H. W. Berg and M. Akiyoshi (*Food Res.*, 1960, **25**, 183—189).—In Zinfandel musts although SO_2 increases colour extraction, the colour stability is inversely proportional to the concn. of SO_2 . Colour stability is a varietal characteristic and Zinfandel musts were the least stable of varieties tested. The effect of SO_2 is different in fermenting musts from that in non-fermenting musts and fermentation is largely responsible for colour losses. A new procedure is suggested for producing dry red and port wines. E. M. J.

Sulphur dioxide and sulphate ions in wines and grape juice. Different methods for their quantitative determination. Method for the simultaneous determination of these compounds by iodometry and gravimetry. L. Deibner (*Rev. Ferment.*, 1959, **14**, 227—244).—Known methods of determination of SO_2 and SO_4^{2-} are compared and discussed. Techniques employed by Deibner and Bénard for determination of SO_2 by iodometry and of SO_4^{2-} by gravimetry, after the formation of $BaSO_4$, are described. (166 references.) J. V. RUSSO.

Flor sherry production by submerged culture. C. S. Ough and M. A. Amerine (*Food Technol.*, 1960, **14**, 155—159).—Conditions of production, e.g., aeration, alcoholic content of the medium, effects on the yeast population of initially starting an aldehyde fermentation and flor sheries aged in glass for 1 year and in small oak cooperage

for 9 months and for 1 year were critically examined. Alcohol content should be kept below 15% by vol. Correlation between alcohol content and max. aldehyde produced was -0.788 , highly significant for the range studied. Delay in starting an initial aerobic fermentation indicates that the cause is an acclimatisation period, not establishment of a mutant yeast strain. Ageing in oak cooperage produced for sherries judged of better quality than the same wines aged in glass. (12 references.) E. M. J.

Clarification of fermented liquor. Aktieselskabet Grindstevaerket (Inventor: K. J. S. Villadsen) (B.P. 812,402, 25.5.54).—Grape juice, wine must, fermented must or fermented wine is clarified by adding at 25–30° a pectolytic enzyme complex capable of precipitating chlorophyll from aq. solution at pH 3–4.5 (20 g. per 100 kg.), then after 40 hr. (in the case of grape juice) or 2–3 weeks (in the case of fermented must or wine) decanting and centrifuging. F. R. BASFORD.

Fruits, Vegetables, etc.

Acid content of apples during the freezing process. IV. Characterisation of damage caused by cold storage of apples especially under freezing conditions. L. Nicolaisen-Scupin (*Källetechnik*, 1960, 12, 50–52).—The determination of titratable total acid content in fruit being difficult, results are subject to fluctuation and therefore to criticism. The use of larger average samples (5–10 apples) tends to eliminate error. Observations show that acidity increases during refrigeration at -14° ; that the increase is less at -5° ; there is an actual increase in acid content, but this does not depend on loss of water from cell contents. An increase in acidity is only detectable in the case of still frozen fruit; during thawing, the acidity falls below that found in fresh fruit. W. H. KEMP.

Carotenoids of apricots. A. L. Curl (*Food Res.*, 1960, 25, 190–196).—The carotenoids were separated by countercurrent distribution into fractions totalling, e.g., hydrocarbons 88, monols 6, diols 3%, and polyols, followed by chromatography to separate the individual constituents. Of the total carotenoids β -carotene amounted to $\sim 60\%$; phytoene, phytyluene, γ -carotene, lycopene, cryptoxanthin and lutein each $\sim 2\%$; 18 other constituents were present in smaller amounts. γ -Carotene and lycopene were recovered in part as polycis isomers, including pro- γ -carotene. Isomerisation of 5,6-epoxide–5,8-epoxide occurred during the ripening. (13 references.) E. M. J.

Composition of cherry seed and cherry seed oil (*Prunus cerasus*). K. G. Weckel and H. D. Lee (*Food Technol.*, 1960, 14, 151–154).—Data are given on the composition of cherry seeds and of cherry kernel oil. The seeds contain protein and oil which may have potential in animal and food mixtures. The kernel oil is similar to that of maize oil, groundnut oil and sesame oil. Elaeostearic acid present may be the cause of accelerated development of oxidative rancidity observed in the oil. (20 references.) E. M. J.

Softening of brined cherries by polygalacturonase and the inhibition of polygalacturonase in model systems by alkylarylsulphonates. W. F. Steele and H. Y. Yang (*Food Technol.*, 1960, 14, 121–126).—The literature is reviewed. Micro-organisms isolated from "cats claw"-infected cherries produced a polygalacturonase and were identified as *Rhizopus nigricans*, *Aspergillus niger*, *Saccharomyces niger*, *Penicillium*, etc. No mould growth was observed on the infected cherries; if the polygalacturonase is of microbial origin it is probably produced by a yeast. The alkylarylsulphonate detergent Naccalon NR was an effective inhibitor for the polygalacturonase in Pectinol M in cherry brine at pH 2.6 and in buffer solutions at pH 3.0, 3.6 and 4.0. (25 references.) E. M. J.

Effects of sugars on the breakdown of pelargonidin-3-glucoside in model systems at 90°. I. J. Tinsley and A. H. Bochian (*Food Res.*, 1960, 25, 161–173).—The variables studied were pH, sugar concn., and amino-acid reaction. In systems buffered at pH 3.4, the rate of pigment breakdown in presence of sugars seemed to be associated with the rate at which the sugar degenerated into furfural-type compounds. Increase in concn. of glucose, fructose or sucrose increased rate of pigment degradation. The degradation of pelargonidin-3-glucoside (I) the chief anthocyanin of strawberries is influenced by the effect of H^+ concn. on the equilibrium between the pseudo-base and the flavylium salt form of the anthocyanin. A rapid degradation of I was observed in presence of furfural and 5-hydroxymethylfurfural. A system containing pigment, sucrose, amino-acid and ascorbic acid at concn. similar to those observed in a sample of strawberry juice gave a rate of pigment degradation almost equal to that observed in the natural strawberry juice sample. (17 references.) E. M. J.

Ripening of bananas. W. Flechtenmacher (*Källetechnik*, 1960, 12, 44–49).—A review of the treatment of bananas during the various phases from harvesting to the last stage of the ripening process. Theoretical considerations, as well as experimental results obtained in the laboratory and under practical conditions are presented. Importance is ascribed to the process of respiration of the fruit, and the effect of R.H. on its wt. and the parts played by ethylene and ozone in the ripening process are discussed. Whilst the former accelerates ripening, the effect is nullified by the safety precautions and food law regulations; the latter has apparently no effect. (10 references.) W. H. KEMP.

Comminuted orange. Novel process for its manufacture. J. B. S. Braverman and A. Levi (*Food Technol.*, 1960, 14, 106–109).—The process was tried out on laboratory and Henze pilot-plant disintegrator scale. Whole oranges are heated in the pressure cooker provided with a blow-out valve at the base where the contents, completely disintegrated, are blown out and the product is subsequently finished and milled. The product is completely sterilised, disintegrated and enzymes are inactivated. E. M. J.

Domestic dates. I. Methods for evaluating darkening. V. P. Maier and F. H. Schiller (*Food Technol.*, 1960, 14, 139–142).—(a) A reflectance method in which deteriorative darkening in whole or pureed dates may be followed is described, the use of pureed dates being preferred. (b) A solvent extraction method permits precise measurement of the amount of sol. date pigment present in date tissue at any degree of darkness. This method has greater advantages in the later stage of darkening. (13 references.) E. M. J.

Rapid method for the determination of sulphur dioxide in sulphited pre-peeled potatoes. L. R. Ross and R. H. Treadway (*Amer. Potato J.*, 1960, 37, 102–107).—The method is based on extraction of cut potatoes with a citrate-phosphate buffer (pH 4.4), which reduces the oxidation of SO_2^{2-} , followed by titration with standard I solution. A. H. CORNFIELD.

Change in the physical condition of starch of the potato during precooking heating. A. L. Potter, E. M. Neel, R. M. Reeve and C. E. Hendel (*Amer. Potato J.*, 1959, 36, 444–449).—Potato starch became less sol. when heated at 60–75°. Solubility decreased with time of heating, but after 60 min. there was little further decrease. This pre-heating resulted in less dissolution of starch from potatoes cooked at 100° for 20 min. The pre-heating before cooking during the prep. of dehydrated mashed potatoes, which has been shown to improve the quality of the product, probably leads to the reduction in the solubility of starch. A. H. CORNFIELD.

Effect of wet soil and carbon dioxide on potato chip colour and sugar content. L. J. Kushman, M. W. Hoover and F. L. Haynes (*Amer. Potato J.*, 1959, 36, 450–456).—Chips made from potatoes grown at high moisture content had a poor colour in every case where the potatoes were stored prior to being processed and in some cases when processed immediately after harvesting. The CO_2 content of the tubers increased with soil moisture level. Holding normal tubers in an atm. of CO_2 also had a bad effect on chip colour quality. The contents of sucrose and reducing sugars were not related to soil moisture, CO_2 content of the atm. during storage, or to chip colour quality. A. H. CORNFIELD.

Microscopical structure of potato chips. R. M. Reeve and E. M. Neel (*Amer. Potato J.*, 1960, 37, 45–52).—Results are presented and discussed. A. H. CORNFIELD.

Bound form of ascorbic acid. XII. Isolation of pure ascorbigen and some other indole derivatives from Savoy cabbage. Z. Procházka and V. Šanda (*Coll. Trav. chim. Tchecosl.*, 1960, 25, 270–280).—The conventional methods for ascorbigen extraction and isolation from fresh Savoy cabbage juice (by Et acetate extraction, concn., dilution with excess of light petroleum—ascorbigen is precipitated, other indolic substances remain in solution—and subsequent fractional pptn. from Et acetate concentrates by non-polar solvents) were complemented by adopting preparative paper chromatography, a countercurrent distribution method and addition of neutral water to the Et acetate extractant. Prep. containing 50% bound ascorbic acid were obtained. Ascorbigen being very unstable, was not obtained in the pure state; indirect methods revealed a composition of $C_{17}H_{17}NO_7$, and a cryst. picrate (m.p. 130°) was prepared. Other indolic substances: indole-3-ylacetoneitrile, indole-3-carboxylic acid and indole-3-aldehyde were isolated from the petroleum filtrate. "Substance C," closely related to ascorbigen (dil. alkali converts ascorbigen into "substance C"), was also isolated, from fractions containing material more hydrophilic than ascorbigen, as an amorphous, white substance. (14 references.) M. LAPIDOT.

Factors affecting decay of prepackaged spinach. R. F. Becker, R. N. Goodman and H. S. Goldberg (*Food Technol.*, 1960, 14, 127–130).—Rate of decay appears to depend on the bacterial population

of the spinach. N-content of the leaves also affects the decay rate. Antibiotics used at low concn. in practical applications would be particularly effective in reducing the no. of bacteria during warm weather, when development of soft rot is most rapid. (14 references.) E. M. J.

Non-alcoholic beverages

Methyl alcohol in fruit juices. B. Weger (*Fruchtsaft-Industrie*, 1960, **5**, 62—68).—The fact that clarified juices normally contain a higher % of CH_3OH than cloudy juices is discussed. Whether the use of clarifying enzymes plays an important part and whether it is advisable to reduce the CH_3OH content by physical means are also discussed. No advantage of one kind of juice over the other could be discovered. As the intake of pectic substances from fruits and vegetables is at least as great as that from the corresponding juice, and as the CH_3OH is excreted with the urine, it makes little difference whether the juice is clarified or not. Nutrition experts are invited to express their views on the physiological aspects of this problem. (17 references.) I. DICKINSON.

Ascorbic acid studies on chilled, fresh and fermented orange juice. M. P. Lamden, C. E. Schweiker and H. B. Pierce (*Food Res.*, 1960, **25**, 197—202).—A survey of the ascorbic acid content of orange juice products showed that juice distributed in cartons and that in retail stores had lower ascorbic acid content than juice of freshly squeezed oranges bought at the same time. In samples containing low amounts of reduced ascorbic acid (57% of the value for fresh orange juice) the amounts of dehydroascorbic acid and diketogulonic acid were relatively high. In fresh orange juice at 4° ascorbic acid content decreased 25% during 4 weeks; substantial increase in oxidised forms was noted 2 weeks later. Fermentation with yeast or bubbling CO_2 through juice in presence of iodoacetate enhanced the stability of ascorbic acid in juice at room temp. E. M. J.

Stability of ascorbic acid in frozen and bottled acerola juice alone and combined with other fruit juices. K. O. Fitting and C. D. Miller (*Food Res.*, 1960, **25**, 203—210).—Acerola (*Malpighia punicifolia*) contains ~2000 mg./100 g. of ascorbic acid. Juices containing approx. 1200 mg. of ascorbic acid per 100 ml. were prepared, frozen and bottled, and were used unsweetened to enhance or fortify pineapple, passion fruit and guava juices which were then frozen or bottled. After storage for 8 months all the frozen juices retained similar proportions of ascorbic acid 82—92%. All bottled juices retained consistently less ~60—70%. Acerola juice had little or no effect on colour or flavour and sugar seemed to have some stabilising effect on colour of frozen acerola juice. E. M. J.

Media for counting sugar tolerant yeasts in concentrated orange juice. M. Ingram (*J. appl. Bact.*, 1959, **22**, 234—247).—Dextrose agar (I), potato I and glucose-citric acid-tryptone agar (II) media are compared. II is troublesome to prepare; colonies grow slowly and are translucent but these characteristics are the result of the high sugar concn. on which the selectivity of the medium depends. Potato I recorded sugar intolerant yeasts; these probably arose from a dirty machine or through faulty canning; I additionally showed bacterial growth. (20 references.) C. V.

Metallic components of fruit juices. III. Oxidation and stability of ascorbic acid in model systems resembling blackcurrant juice. IV. Oxidation and stability of ascorbic acid in blackcurrant juice. C. F. Timberlake (*J. Sci. Fd Agric.*, 1960, **11**, 258—268, 268—273).—III. The systems were of pH 2.9 and consisted mainly of solutions of ascorbic acid in partly-neutralised citric acid with concn. approximating those of the juice. O_2 consumptions were measured by the Warburg technique and ascorbic acid losses were followed concurrently. In presence of Cu (0.85 p.p.m.) traces of Fe^{2+} markedly increased the oxidation of ascorbic acid over that found with Cu alone. Blackcurrant anthocyanins increased O_2 uptake and ascorbic acid loss, but glucose in presence of Cu alone reduced reaction rate. Ascorbic acid losses in presence of Cu with and without Fe increased according to buffer solutions in the order: malonate citrate, malate, quinate, phthalate, formate and phosphate. The effects of metal chelating agents are described. (55 references.)

IV. In the natural uncontaminated blackcurrant juice results indicate that oxidation of ascorbic acid in presence of air is a non-enzymic mechanism, catalysed by natural Cu and enhanced by natural Fe and anthocyanins present. Contamination by Cu (e.g., from spray residues) greatly increased oxidation of ascorbic acid. The action of chelating agents (e.g., ethylenediaminetetra-acetate) is discussed. E. M. J.

Production of concentrates of foodstuffs. I. A. M. Burger (*Riechstoffe u. Aromen*, 1960, **10**, 80—85).—Industrial production of citrus and other fruit concentrates is described.

H. L. WHITEHEAD.

Use of cation-exchange capacity of citrus juices as rapid screening test for detection of adulterations. Y. Pomeranz, J. J. Monselise and C. Lindner (*Bull. Res. Council Israel*, 1959, **7C**, 171—174).—The test is based on the difference (Δ , ml. n-NaOH per 100 ml. juice) between a direct acidimetric titration and the titration made after cation-exchange with H^+ on a 30-cm. column of Duolite C 20. Suggested lower permissible limit of Δ is 4.5 for genuine Jaffa-orange juice; the ash/ Δ values are approx. constant.

W. J. BAKER.

Tea, coffee, cocoa

Fractionation of nitrogenous components in black tea. C. P. Natarajan, N. Chandrasekhara and D. S. Bhatia (*Food Sci., Mysore*, 1959, **8**, 390—391).—Tea contains about 3.3% of non-caffeine N. Extraction of powdered tea with different solvents showed that it contained 0.6% of protein sol. in water, 10.0% sol. in 1% NaOH, and 1.1% sol. in 80% ethanol (prolamines). There was no protein sol. in 10% NaCl. Of the protein sol. in NaOH, 39% was precipitated by trichloroacetic acid, and 78% by phosphotungstic acid.

M. D. ANDERSON.

Rheometry and rheology of molten chocolate. III. Calculations on flow in pipes. A. Fincke and W. Heinz (*Fette Seif. Anstrichm.*, 1960, **62**, 197—204).—A general formula is derived from which it is possible to calculate the flow of molten chocolate through pipes, provided that the pipe dimensions, pumping pressure and the rheological properties of the chocolate are known. Some theoretical results have been experimentally verified. G. R. WHALLEY.

Milk, Dairy Products, Eggs

Strontium in milk. I. Removal by means of reverse-flow ion-exchange columns. II. Removal by batch ion-exchange methods. D. G. Easterly, B. J. Demott and R. G. Cragle (*J. Dairy Sci.*, 1960, **43**, 137—145, 146—150).—I. Between 70 and 80% of the ^{89}Sr and ^{90}Sr was removed from the second 100 ml. and 50% from the tenth 100 ml. of skim milk from cows given these isotopes orally by upward-flow ion-exchange resin column packed with either Dowex 50W-X12 or Dowex 50W-X4; the Ca form of the resins was more effective than was the Na form. Duolite C-20 resin was ineffective. Recoveries were similar when the isotopes were added directly to the skim milk and allowed to stand for 16 hr., but less ^{90}Sr was removed. Preferential removal of Sr over Ca occurred. Treated milk had a higher Ca content, lower pH, greater titratable acidity and shorter rennet coagulation time than had untreated milk.

II. When skim milk containing ^{89}Sr and ^{90}Sr was in contact with Dowex 50W-X12 (in the Ca form) apparent equilibrium was reached in 1 to 10 min., and the removal of ^{89}Sr and ^{90}Sr varied from 93 and 94% respectively with a milk/resin ratio of 2:1 to 21 and 16% respectively with a ratio of 200:1. With the former ratio, 4.27% of the resin capacity was used for exchange of Sr and Ca, and with the latter ratio, 73.6%. S. C. JOLLY.

Method for the elimination of ashing in strontium-90 determination of milk. G. K. Murthy, J. E. Coakley and J. E. Campbell (*J. Dairy Sci.*, 1960, **43**, 151—154).—After adding Sr carrier to the milk sample and filtering off the protein precipitated by an equal vol. of trichloroacetic acid (24%), the alkaline earths in the filtrate are precipitated by adjusting the pH to 8.5—9.0 with NaOH in the presence of Na_2CO_3 and the ppt. is analysed for ^{90}Sr as described by Murthy *et al.* (*ibid.*, 1959, **42**, 1276) for milk ash. The results are not significantly different from those obtained on milk ash.

S. C. JOLLY.

Solubility of tricalcium citrate in solutions of variable ionic strength and in milk ultrafiltrates. M. Boulet and J. R. Marier (*J. Dairy Sci.*, 1960, **43**, 155—164).—The solubility product of tricalcium citrate in solution at equilibrium varied with ionic strength (μ), according to the relation $\text{p}K_s = 17.63 - 10.84\sqrt{\mu}$, but was unaffected by pH in the range 4.4—8.8, temp. in the range 21° to 95°, or by the presence of Mg^{2+} or PO_4^{3-} . S. C. JOLLY.

Ultracentrifugation studies of milk heated to sterilisation temperatures. H. K. Wilson, E. O. Herreid and R. McL. Whitney (*J. Dairy Sci.*, 1960, **43**, 165—174).—Ultra-high-temp. short-time heating of milk increased the sedimentation of protein particles on ultracentrifugation, the sediment vol. increasing with increasing severity of heating. After storage for 72 days at 40°F the max. vol. of the sediment was ~10% greater, but at 70° and 100°F it was less than it was initially, indicating dissociation of the larger particles during storage. S. C. JOLLY.

Quantitative test for carrageenin ester sulphate in milk products. P. M. T. Hansen and R. McL. Whitney (*J. Dairy Sci.*, 1960, **43**, 175—186).—A gravimetric method, based on the determination of sulphate after preliminary dialysis and hydrolysis, is described for

the determination of carrageenin in milk products. When applied to the assay of ester-sulphate contents of various salts of κ - and λ -carrageenin the results with λ -carrageenin were in agreement with the commonly accepted structure of this fraction, but those with κ -carrageenin indicated the need for slight modification to its reported structure. If the hydrolysis step is omitted, the method could be used to determine sulphate in milk. S. C. JOLLY.

Role of leucocytes in the reduction of resazurin in raw milk. J. J. R. Campbell and R. A. Phelps (*J. Dairy Sci.*, 1960, **43**, 187—192).—When freshly isolated from bovine blood, leucocytes rapidly reduced resazurin in raw milk, but their activity quickly disappeared on storage in milk at 37°. Leucocytes isolated from raw milk had little reducing activity, and their removal by centrifugation had little effect on dye reduction by the milk. Incubation of the leucocytes isolated from raw milk with fresh bovine plasma increased their reducing activity. Disintegrated leucocytes do not provide substrates for the reducing systems of raw milk. S. C. JOLLY.

Xanthine oxidase and incidence of spontaneous oxidised flavour in milk. G. J. Smith and W. L. Dunkley (*J. Dairy Sci.*, 1960, **43**, 278—280).—Determination of xanthine oxidase activity and the incidence of spontaneously developed oxidised flavour did not confirm the findings of Aurand and Woods (*ibid.*, 1959, **42**, 1111) of good correlation between the properties. Spontaneous oxidised flavour may not be differentiated from non-spontaneous flavour on the basis of catalytic activity of xanthine oxidase. Although this activity may be involved, it is not a limiting factor. S. C. JOLLY.

Keeping quality of pasteurised Grade A milk offered for sale in the Chicago market. L. D. Witter, P. H. Tracy and H. K. Wilson (*Ill. agric. Exp. Sta.*, 1959, Bull. 646, 20 pp.).—Effects of period and temp. of storage on pasteurised milks are recorded. Detectable changes in milk quality may be expected after 6 days' storage at 55°F. A. G. POLLARD.

Milk substitutes of vegetable origin. I. Nutritive value of milk substitutes prepared from soya-bean and groundnut. S. R. Shurpalekar, N. L. Lahiry, M. R. Chandrasekhara, M. Swaminathan, K. Indiramma and V. Subrahmanyam (*Ann. Biochem.*, 1959, **19**, 269—274).—Preliminary studies on the overall nutritive value of soya-bean and groundnut milks and the protein-efficiency ratio of the proteins of spray-dried powder obtained from a blend of soya-bean and groundnut milks were assessed by feeding tests on rats. The protein efficiency ratio (I) of the proteins (at 10% level) of spray-dried vegetable milk powder (from a blend containing 2 pt. soya-bean milk and 1 pt. groundnut milk) and of skim milk powder were 1.93 and 3.21 (4-week period) and 1.68 and 2.28 (8-week period) respectively. The I of proteins of a mixture of 4 pt. of vegetable milk powder and 1 pt. skim milk powder were 2.49 and 2.02, respectively, in the same periods. J. V. RUSSO.

Multiplication of coli-aerogenes bacteria in milk stored at 3—5°. J. J. Panes and S. B. Thomas (*J. appl. Bact.*, 1959, **22**, 272—277).—Samples (108) were held for 72 hr. at 3—5°; 35.2% showed an increase in coli-aerogenes organisms of over 100-fold while 10.2% increased over 1000-fold. The predominating organisms were *Klebsiella cloacae* and *K. aerogenes* 1. Some strains showed scanty and slow growth on yeast dextrose agar but most strains grew luxuriantly; these latter are considered to be typical facultative psychrophiles of milk. C. V.

Analysis of the performance of an ultra-high-temperature milk sterilising plant. IV. Comparison of experimental and calculated sporicidal effects for a strain of *Bacillus stearothermophilus*. H. Burton, J. G. Franklin, D. J. Williams, H. R. Chapman, A. Jean, W. Harrison and L. F. L. Clegg (*J. Dairy Res.*, 1959, **26**, 221—226).—The sporicidal performance against highly resistant spores of the thermophile *B. stearothermophilus* of an ultra-high-temp. sterilising plant calculated from time and temp. distributions in the plant and laboratory bacteriological data agreed well with that from direct plant experiments. Interpretation of the results was complicated by the very marked inhibition of the organism by treated milk. Because of the uncertainties as to the possible no. and types of microorganisms in milk, this theoretical method of estimating performance is unlikely to give reliable information on spore age under practical dairy conditions. S. C. JOLLY.

Coconut-like flavour defect from milk fat. III. Origin of δ -decalactone in fat-containing dairy products. L. R. Mattick, S. Patton and P. G. Keeney (*J. Dairy Sci.*, 1959, **42**, 791—798).—The precursor of the compound, δ -decalactone, responsible for the coconut-like flavour which may develop in fat-containing dairy products is probably 5-hydroxydecanoic acid present in butterfat as a simple ester. On low-temp. crystallisation of butterfat from acetone, this ester was concentrated in the low-melting fraction (—50° filtrate)

and was further resolved by chromatography on alumina into a fraction representing ~0.4% of the original fat. S. C. JOLLY.

Inhibition of lipase and lipolysis in milk by *N*-ethylmaleimide. N. P. Tarassuk and M. Yaguchi (*J. Dairy Sci.*, 1959, **42**, 864—865).—Addition of *N*-ethylmaleimide (0.02M) to milk inhibited lipase activity and, to a smaller extent, level of induced lipolysis, possibly because of the protective effect of the fat-globule membrane. Thus, —SH groups are essential for both induced and spontaneous lipolysis, and are probably involved in the formation of the specific lipase-substrate complex. The active sites of the lipase are at or near the —SH groups. S. C. JOLLY.

Milk powders. II. Method for the estimation of the wettability of milk powders. B. E. Baker and E. Bertok. **III. Preparation and properties of milk powders containing low-melting butter oil.** B. E. Baker, E. Bertok and E. R. Samuels. (*J. Dairy Sci.*, 1959, **42**, 869—872, 1038—1044).—II. A modification is described of the method of Kleinert (*Schweiz. Milchztg.*, 1949, **75**, 379; *Chem. Abstr.*, 1950, **44**, 6538) which allows small differences in the wettability of milk powders to be detected. Generally, the wettability and, to a lesser degree, the dispersibility of whole-milk and skim-milk powders decrease with decreasing particle size of the powders.

III. The wettability and dispersibility of milk powder was affected by the m.p. of the fat in the powder; those of a powder containing ~25% of a butterfat fraction melting at 19° to 21° were approx. equal to those of a commercial "instant" skim milk powder. S. C. JOLLY.

Some factors involved in the development of oxidised flavour in milk. L. W. Aurand, A. E. Woods and W. M. Roberts (*J. Dairy Sci.*, 1959, **42**, 961—968).—An enzyme mechanism is responsible for the production of spontaneous oxidised flavour in milk, whereas a chemical oxidation is involved in the induced oxidised flavour. The former flavour defect is prevented either by heating or by the use of an enzyme inhibitor (*p*-chloromercuribenzoate), but these treatments have no effect on Cu-induced oxidised flavour, which is prevented by the use of a chelating agent (2,9-dimethyl-1,10-phenanthroline) and by cysteine. Enzyme-inhibited milk is re-activated by cysteine, thus suggesting that an enzyme is involved in producing spontaneous oxidised flavour in milk, and that an —SH group is essential at the active site of the enzyme for activity. S. C. JOLLY.

Effect of thermal oxidative polymerisation on the growth-promoting value of some fractions of butterfat. V. R. Bhalerao, O. C. Johnson and F. A. Kummerow (*J. Dairy Sci.*, 1959, **42**, 1057—1062).—Data from growth experiments with rats indicated that butterfat contains acetone-insol. triglycerides which either counteract the toxic effects of products formed during thermal polymerisation of the fat or prevent their formation. S. C. JOLLY.

Adhesion-cohesion, static friction and macrostructure of butters. I. Method of measuring adhesion-cohesion. J. W. Claassens (*S. Afr. J. agric. Sci.*, 1958, **1**, 457—463).—A modified Westphal sp. gr. balance is described by means of which a vertical pull is applied to measure the force with which butter adheres to solid materials ("stickiness"). From the counter-wt. required to detach the adhering body, the force ofhesion (analogous to sorption) may be determined in dynes per cm.² (9 references.) S. C. JOLLY.

Slow and fast acid-producing variants of strains of *Streptococcus cremoris* and *Str. lactis* used as cheese starters. I. Garvie (*J. Dairy Res.*, 1959, **26**, 227—237).—Growth of *Str. cremoris* 924 on milk agar was improved in the presence of CO₂. When slow and fast acid-producing variants were mixed and subcultured daily, all the resulting cultures were active acid producers, irrespective of the initial ratio of the slow and fast variants. If the strains are correctly selected, single-strain starter cultures can be maintained apparently indefinitely by daily subculture in litmus milk and by freeze drying. Activity varies widely, and is apparently a function of the culture. S. C. JOLLY.

Viscosity changes in concentrated skim milk treated with alkali, urea and calcium-complexing agents. I. Importance of the casein micelle. R. Beeby and K. Kumet. **II. Influence of concentration, temperature and rate of shear.** R. Beeby and J. W. Lee (*J. Dairy Res.*, 1959, **26**, 248—257, 258—264).—I. Addition of urea, Ca-complexing agents or alkali to conc. skim milk destroyed the characteristic opacity of the milk and markedly changed the η , the η -time curve passing through a distinct max. within a short time. These changes are attributable possibly to expansion of the casein micelles, probably due to electrostatic repulsion within the micelle, followed by disintegration into "molecular" units. Ca, either in the ionic atm. of the micelle or as Ca bridges within the micelle, and H bonds are the major factors responsible for maintaining the stable structure of the casein micelle.

II. An essentially exponential relation between max. η and concn. of casein existed for conc. skim milk treated with Ca(OH)₂. Positive

temp. coeff. occurred with samples treated with alkali and negative coeff. with those treated with Na polymetaphosphate. The time of appearance of the max. was independent of temp. with samples treated with NaOH or Ca(OH)₂, but when Na polymetaphosphate was used the max. appeared earlier at higher temp. The η of all systems decreased with increasing rates of shear, the deviation from Newtonian being greatest when η was at its max. The effects are consistent with the swelling and bursting of casein micelles postulated in part I.

S. C. JOLLY.

Cream paste manufacture. R. Lees (*Confect. prod.*, 1960, **26**, 227, 229—230).—A brief review; possible faults, and their prevention, are discussed.

C. V.

Vitamin B₁₂ in wheys prepared by coagulating milk with acid and rennet. E. B. Collins and M. Yaguchi (*J. Dairy Sci.*, 1959, **42**, 1927—1931).—The average vitamin B₁₂ contents of three samples of pasteurised milk and 16 samples of pasteurised skim milk were 4.0 and 4.7 $\mu\text{g.}$ per l. respectively. Both casein and whey proteins bound the vitamin. Rennet released some of the bound vitamin from casein, the amount depending on the rennet concn. and the time at 40°. On average, 60% of the vitamin present in the original milk was in six renneted wheys and 44% in six acid wheys.

S. C. JOLLY.

Commercial enzymes by extraction: rennet. C. Placek, V. S. Bavisotto and E. C. Jadd (*Industr. Engng Chem.*, 1960, **52**, 2—8).—Production from calves' stomachs is described. O. M. WHITTON.

Butter manufacture. L. Eisenreich (*Fette Seif. Anstrichm.*, 1960, **62**, 295—297).—Present-day practice in the continuous process for butter manufacture is discussed.

J. L. PROSSER.

Butter technology. W. Mohr (*Fette Seif. Anstrichm.*, 1960, **62**, 285—292).—Progress in the elucidation of formation, structure and texture of butter, and in plant for its manufacture, is reviewed.

J. L. PROSSER.

Some volatile compounds in New Zealand and Cheddar cheese and their possible significance in flavour formation. I. Identification of the volatile carbonyl fraction. J. R. L. Walker and R. J. Harvey. **II. Volatile compounds of sulphur.** J. R. L. Walker (*J. Dairy Res.*, 1959, **26**, 265—272, 273—276).—I. The following carbonyl compounds were detected by paper chromatography and other methods in steam distillates from mature N.Z. Cheddar cheese (approx. concn. in p.p.m. in the cheese are given in parentheses): acetoin, diacetyl, acetaldehyde (2.65), acetone (0.86), butan-2-one (0.80), pentan-2-one (2.47), heptan-2-one (6.60), nonan-2-one (1.70) and undecan-2-one (0.83). The possible rôle of these compounds in the typical flavour of the cheese is discussed.

II. H₂S was the only volatile S compound detected in gas-streams or steam-distillates from mature cheese. After removal of this and the carbonyl compounds, the original cheesy aroma of the gas-streams and distillates was completely removed. The rôles of H₂S and the carbonyl compounds in Cheddar cheese flavour are discussed.

S. C. JOLLY.

Use of an oxidation-reduction indicator to compare the oxygen permeabilities of films for rindless cheese. R. M. Dolby (*J. Dairy Res.*, 1959, **26**, 281—283).—The extent of bleaching after 14 days of the methylene blue used to colour the surface of slices and blocks of cheese vacuum-packed or wrapped in film gave a good indication of the extent to which O₂ was excluded by the film. Similar results were obtained with vacuum-packed slices and with wrapped rindless cheeses. With polythene-coated and wax-coated MSAT Cellophane films there was little or no reduction of the dye on the cheese surface. Similar films, including a lamination of foil or MXXT Cellophane, allowed complete or almost complete bleaching.

S. C. JOLLY.

Preparation of calcium-sensitive α -casein. C. A. Zittle, J. Cerebulis, L. Pepper and E. S. Della Monica (*J. Dairy Sci.*, 1959, **42**, 1897—1902).—A relatively simple modification of the method of fractionating whole casein in urea solution is described which gives a Ca-sensitive α -casein (I) in approx. 40% yield of the total casein. I appeared to be homogeneous on electrophoresis at pH 2.3 and 8.5, and very little (<1%) was solubilised by rennin. I and α -paracasein were distinguished by their sensitivities to low concn. of Ca²⁺ (1.5 to 3.5 mM per l.) and the initial courses of their aggregation in the presence of Ca²⁺.

S. C. JOLLY.

Some esterases in cows' milk. T. L. Forster, H. A. Bendixen and M. W. Montgomery (*J. Dairy Sci.*, 1959, **42**, 1903—1912).—Experiments on the effects of organophosphorus enzyme inhibitors (Isopetox, parathion, Dyflos and ethyl pyrophosphate) on the activity of milk enzymes at pH 8.0 at 37° towards different substrates indicated the presence of three distinct esterases in raw milk. One was responsible for ~95% of the activity of milk towards tributyrin, benzyl benzoate and the stearoyl and butyryl esters of sodium 2-hydroxynaphthalene-6-sulphonate, the second for 60 to 70% of

the activity towards phenyl propionate, and the third for ~80% of the activity towards phenyl acetate. Each esterase appeared to hydrolyse at least one of the other substrates.

S. C. JOLLY.

Total monoglyceride content of some dairy products. R. G. Jensen, G. W. Gander and A. H. Duthie (*J. Dairy Sci.*, 1959, **42**, 1913—1916).—The average total monoglyceride content (as mmoles per 100 g. of fat) of fresh raw milk, pasteurised whole milk, homogenised milk, cream (40% fat), butter and Blue cheese was 0.077, 0.145, 0.206, 0.119, 0.189 and 0.354, respectively. Some implications of these contents are discussed.

S. C. JOLLY.

Inhibition of certain types of bacterial spoilage in creamed cottage cheese by the use of a creaming mixture prepared with *Streptococcus citreovorus*. D. W. Mather and F. J. Babel (*J. Dairy Sci.*, 1959, **42**, 1917—1926).—Slime formation on creamed cottage cheese inoculated with *Pseudomonas* spp. was prevented by a special creaming mixture consisting of 1.5 parts of cream (20% fat) and 1 part of an acidified culture of *Streptococcus citreovorus*; coliforms were also inhibited, but *Geotrichum candidum* and *Candida pseudotropicalis* were not. The reduction in the diacetyl content of normally creamed cheese by the *Pseudomonas* spp. did not occur with the special creaming mixture. Slime formation was not prevented so effectively by the addition of acetic or propionic acid to the normally creamed cheese or by reducing the pH to 5.2. Neither volatile acids nor pH was responsible, therefore, for the inhibition.

S. C. JOLLY.

Recent developments in cottage cheese manufacturing procedures. W. V. Price, A. M. Swanson and D. B. Emmons (*J. Dairy Sci.*, 1959, **42**, 2005—2008).—A survey.

S. C. JOLLY.

Cottage cheese cultures. F. J. Babel (*J. Dairy Sci.*, 1959, **42**, 2009—2011).—A survey.

S. C. JOLLY.

Factors affecting the yield of cottage cheese. W. A. Cordes (*J. Dairy Sci.*, 1959, **42**, 2012—2015).—A survey.

S. C. JOLLY.

Keeping quality of cottage cheese. N. C. Angevine (*J. Dairy Sci.*, 1959, **42**, 2015—2020).—A survey.

S. C. JOLLY.

Edible Oils and Fats

Heat transfer in hot fat cooking. H. L. Smith, jun. (*Food Technol.*, 1960, **14**, 84—88).—The Smitherm method of indirect heating of cooking fats is described. The heat transfer or absorbing surface in the heat generator is composed of steel tubes or pipes through which is circulated a high-boiling liquid. The advantages of the method are summarised; e.g. reduction in heat damage, because the fat does not come in contact with a high temp. surface, reduction in rate of increase in free fatty acids and precise temp. control of the fat, etc.

E. M. J.

Composition of seed fat of Ceylon sweet orange. A. H. Weerakoon (*J. Sci. Fd Agric.*, 1960, **11**, 273—276).—The fatty oil which forms ~33% of the dried whole seed has the following component fatty acids: palmitic 21.8% wt., oleic 27.4%, linoleic 34.2%, myristic 6.8%, stearic 6.4%, linolenic 2.1%, arachidic 0.6%, lauric 0.6% of the total fatty acids. The constituent glycerides of the oil comprised monosaturated diunsaturated 54% and disaturated monounsaturated ~22%, triunsaturated ~18% and trisaturated glycerides ~6%.

E. M. J.

Oxidation of fat in model systems related to dehydrated foods.

S. J. Bishov, A. S. Henick and R. B. Koch (*Food Res.*, 1960, **25**, 174—182).—Model emulsion systems designed to simulate dehydrated hydrocolloids were prepared and studied under accelerated storage conditions for deteriorative reactions. Phospholipids in concn. of 0.5—5.0% of dry wt. of fat protected against oxidation; protein-containing emulsions were more stable than those containing only oil. The catalytic effect of haemoglobin on acceleration of oxidative reaction resulted in spontaneous combustion in the dried emulsions at 85°. Pro-oxidant effects of ferric citrate were small compared with those of porphyrin compounds. Phospholipids alone or with proteins had significant antioxidant action in dehydrated emulsions containing porphyrin compounds but butylated hydroxyanisole and nordihydroguaiaretic acid (0.03%) of dry wt. of fat, had only slight effect. (16 references.)

E. M. J.

Lipolytic activity of micro-organisms at low and intermediate temperatures. I. Action of *Pseudomonas fluorescens* on lard. J. A. Alford and L. E. Elliott (*Food Res.*, 1960, **25**, 296—303).—The effect of medium composition surface-to-vol. ratio, pH and temp. on the production and activity of lipase by *P. fluorescens* when lard was the substrate was studied. Peptone broth (1%) with initial pH of 7.0 gave max. lipase production, yield being greater in shallow, stationary layers than in deeper layers or in aerated cultures.

Optimum pH was ~7.0 with some production at pH 6.0. At 20° levels of lipase were observed in 2–3 days, production was very slight at 30° although all yields were good. Amount of enzyme produced per cell at 5° equalled that produced at 20°. Optimum temp. for lipase was near 40° and was not affected by temp. at which cells were grown. (13 references.) E. M. J.

Antioxygenic activity of lecithin and its hydrolysis products. III. Lecithin, glycerophosphorylcholine, phosphorylcholine, phosphatidic acids, choline and glycerol. C. Urakami and H. Kameyama (*Bull. chem. Soc. Japan*, 1960, **33**, 29–33).—Purified ovo-lecithin, DL- α -glycerophosphorylcholine, choline and glycerol exhibit no antioxygenic property against carefully purified methyl oleate. Phosphorylcholine, its chloride and natural phosphatidic acids show activity. The order of activity efficiency at 60° is orthophosphoric acid > phosphatidic acid and phosphorylcholine > phosphorylcholine chloride > α -glycerophosphoric acid > β -glycerophosphoric acid. It is suggested that the phosphoryl moiety is predominantly responsible for antioxygenic activity. I. JONES.

Vegetable fat, particularly for a margarine composition. E. F. Drew (B.P. 813,402, 23.5.56).—A fat composition, for use in the production of margarine, comprises a glyceride oil of the coconut group, glycerides of the palmitic group (1–15), and (in physical or chemical combination) glycerides (1–15 wt.-%) of low-mol. fatty acids (e.g., caproic, caprylic and capric acid). If desired, the coconut oil fraction may be employed in hydrogenated form. In an example, coconut oil is heated with mixed glycerol esters (~5) of caproic, caprylic and capric acids until homogeneous, then glycerides of palmitic acid containing >5% of triglycerides of coconut oil fatty acids of 14 C or 18 C or more C are added (6 wt.-%), to give the modified fat product. F. R. BASFORD.

Meat and Poultry

Efficacy of oxytetracycline [OTC] for ageing beef. R. B. Sleeth and H. D. Naumann (*Food Technol.*, 1960, **14**, 98–101).—The effect, under experimental conditions, of, e.g., 30 min. *ante mortem* injection of aq. OTC to control surface and internal microbial growth during ageing was studied. Intramuscular injections in the tail (0.80 mg. of OTC/lb. of body wt.) were administered to control deep-seated spoilage and u.v. radiation to control surface growth when the carcass was aged at 86°F. To avoid development of undesirable appearance caused by the u.v. radiation an external spray of 20 p.p.m. of OTC + 20 p.p.m. nystatin was used. Bacterial, coliform, yeast and mould counts were negligible after carcasses were washed and sprayed with OTC and nystatin prior to ageing. If beef treated with OTC were cooked to 160°F before consumption untoward effects of the antibiotic to public health would be minimised. (14 references.) E. M. J.

Use of antibiotics and γ -irradiation in the ageing of steaks at high temperatures. G. D. Wilson, P. D. Brown, W. R. Chesbro, B. Ginger and C. E. Weir (*Food Technol.*, 1960, **14**, 143–147).—Microbial growth during ageing was effectively controlled by infusion of 30–50 p.p.m. of oxytetracycline into round steaks prior to cutting or by 45,500 rad of γ -radiation combined with antibiotic treatment. At this level, γ -irradiation did not affect tenderness or tenderisation of steaks aged at 110°F for 24 hr. In tests varying temp. and period of ageing the most desirable conditions for rapid tenderisation of beef round steaks were 24 hr. at 110°F. These steaks had tenderness increases comparable to those in steaks aged for 14 days at 35°F. E. M. J.

Degree of muscular contraction as a factor in tenderness of beef. R. H. Locker (*Food Res.*, 1960, **25**, 304–307).—The recognition of fibril patterns, out of four distinct known types, offers a convenient measure of the degree of contraction. The distribution of these four is recorded for various muscles representing a complete range of tenderness. Tasting tests on *psaos* muscles which had been cut at death and allowed to shorten showed they were tougher than controls. Relaxed muscles are tenderer than partly contracted muscles and this effect may be significant in the grading of muscles of low connective tissue content. E. M. J.

Estimation of beef marbling and its relationship to tenderness and juiciness. G. H. Wellington and J. R. Stauffer (*Cornell agric. Exp. Sta.*, 1959, Bull. 941, 30 pp.).—Photographic standards showing 11 degrees of marbling of rib-eye steaks from beef cattle of widely different fat content are presented. Tenderness, as measured by mechanical shear resistance of cooked steaks, was not significantly related to degree of marbling. Although tenderness as estimated by a taste panel was significantly correlated with degree of marbling, the tenderness differences accounted for only 7% of the tenderness variability. Juiciness increased significantly with degree of marbling. A. H. CORNFIELD.

Digestibility of beef from cattle of different ages. R. Grau (*Fleischwirtschaft*, 1960, **12**, 166–168).—*In vitro* experiments were made using pepsin hydrochloride. Comparison was made between back muscle and shin from calves (3–4 weeks), cattle (1–2 years, 5 years, 13–15 years) and frozen meat stored for 2 and 12 months. Digestibility of the finely ground meat was independent of age but that from the shin appeared to be slightly less digestible than that from the back. C. V.

Up-grading of low-grade meat. I. F. Penny (*Chem. & Ind.*, 1960, 288–289).—The accelerated freeze-drying process (*Brit. chem. Engng.*, 1959, **7**, 390) can be applied to low-grade beef before reconstitution in enzyme solution and subsequent tenderising with ficin (0.0025%) or papain (0.005%). Inclusion of NaCl, Na glutamate or Protex in the enzyme solution improves flavour and apparent juiciness. The tenderisers are active only during the “warming-up” period (>90°); cooking time is reduced. W. J. BAKER.

X-Ray examination of frankfurter canned sausages. B. Malčić and S. Rapić (*Fleischwirtschaft*, 1960, **12**, 23–26).—Brine content, bone particles, pre-decomposition, etc. can be recorded but decomposition without gas formation cannot be seen. Since the filling is intermingled with air bubbles, the X-ray gives a fine spongy appearance. It is unnecessary to use film as cheap X-ray paper gives satisfactory results. C. V.

Effect of sugar on the flavour and colour of smoked hams. F. Mills, C. E. Weir and G. D. Wilson (*Food Technol.*, 1960, **14**, 94–97).—If a sweetening agent is desired in rapidly cured hams, sucrose, dextrose or a cyclamate appear to be interchangeable. The threshold level for detection of sweetness is 0.50–0.75% of sugar, corresponding to 50–75 lb. of sugar/100 gal. of pickling brine. Sugar had no effect on colour production or stability. (11 references.) E. M. J.

Influence of low-level γ -irradiation, antibiotic treatment, storage temperature and vacuum packing on flavour and bacterial changes in cured bacon. W. L. Brown and M. L. Schmucker (*Food Technol.*, 1960, **14**, 92–93).—Each of the above-mentioned factors had its effect on the quality of sliced bacon. Low storage temp. (28–32°F) and vac. packing were slightly more effective in maintaining an acceptable product than were the other treatments tested. E. M. J.

The importance of detection of *Streptococcus faecalis* in meat and meat products. F. Kelch and E. Stehle (*Fleischwirtschaft*, 1960, **12**, 92–96).—Swabs (146) from hog carcasses showed presence of faecal streptococci (Group D. Lancefield), *S. faecium* (48, 13 strains) and *S. faecalis* (*liquifaciens*) (63, 10 strains). Hams (175) incubated five days at 37° showed faecal streptococci in 45%, *S. faecium* being present in 90% of these. (26 references.) C. V.

Bactericidal power of the tetrathionates. K. Lang (*Fleischwirtschaft*, 1960, **12**, 292–293).—Comparison of the tetrathionates (I) showed that Na I (0.5%) was effective against normal and resistant strains of *Bact. coli*, *Aerobacter aerogenes*, *Proteus vulgaris*, *Streptococcus faecalis*, *Micrococcus pyogenes* (var. *aureus*), *Erysipelothrix murisepitica* and *Bacillus mesenteroides*. The K salt (II) (0.56%), possessed no bactericidal effect against these organisms while at 1.5% there was little difference; at 2% a degree of activity was noted. In a further series 0.5% of Na I was found to be effective against *Salmonella cholerae* (type America and var. *Kunzendorf*) and *S. London* while II (1.5 and 2%) gave identical results; II in a 0.56% concn. was ineffective. Neither of the I compounds was effective against *S. Dublin*, *S. abortus-equina*, or *S. gallinarum pullorum* at any of the indicated concn. C. V.

Doneness of frozen, defrosted turkey halves roasted to several end point temperatures. G. E. Goertz, K. Cooley, M. C. Ferguson and D. L. Harrison (*Food Technol.*, 1960, **14**, 135–138).—Turkey halves roasted to either 90° in the *pectoralis major* or 95° in the thigh were satisfactorily done, a temp. of 90° in the *pectoralis major* being preferred because of the size and greater uniformity of that muscle. Halves cooked to 85° in the thigh were underdone. Flavour and tenderness scores were similar for birds cooked to all end-point temp. E. M. J.

Bacterial flora of poultry kidneys and effects of kidney removal on yield and shelf-life. E. O. Essary and C. E. Howes (*Poultry Sci.*, 1960, **39**, 56–59).—Yield losses due to kidney removal ranged from 0.66% to 0.82% for 8-week broilers, 10-week fryers and 1-year-old White Leghorns. Kidney removal resulted in slightly greater increase in wt. on chilling in ice slush. Shelf-life of fryers was increased by an average of 1.27 days by kidney removal. A. H. CORNFIELD.

Fish

Composition and amino-acid content of high-grade whale meat meal. H. Pritchard and P. A. Smith (*J. Sci. Fd Agric.*, 1960, **11**, 249–252).—Seven representative samples were obtained: two from

floating factories, two from land stations and three from a floating factory from whales killed at different intervals before processing. Overall composition did not vary widely within the class of No. 1 grade meals, but that produced on the land station contained more vitamin B₁₂ and less pantothenic acid than that produced on the floating factory. Variation in free fatty acid content of the oil is due to length of time which elapses between killing and prep. of the meal. Comparative data on essential amino-acid contents of whale meat meal and other protein meals, e.g., meat and bone meal, white fish meal and whole wheat meal are given. E. M. J.

Correlation of taste panel gradings with salt-extractable protein of frozen fish filets. M. N. Moorjani, W. A. Montgomery and G. G. Coote (*Food Res.*, 1960, **25**, 263—269).—Data, capable of being analysed statistically, related to the ability of a taste panel to detect texture differences in fish held in frozen storage, the amount of extractable actomyosin and the correlation between them were obtained. For three storage conditions: -10° and -18° in heat-sealed M.S.A.T. Cellophane bags and at -18° in cans under vac. (22 in. Hg) differences in salt-extractability were associated with texture differences for storage times of 2—6 months. Two rates of loss of extractability were indicated by the regression of protein on time of storage. Initial rapid rate of change depended on temp.; the subsequent slower rate was the same for the three storage conditions. (10 references.) E. M. J.

Water content of cod (*Gadus callarias* L.) muscle. R. M. Love (*Nature, Lond.*, 1960, **185**, 692).—Analyses were made at different seasons of the water content of the muscle of cod fish (i) up to 20 in. long (immature), (ii) about 30 in. long (reaching maturity), and (iii) 36 to 42 in. long (mature). All showed max. water content in March, extent of hydration increasing with age of fish. The duration of the increase in water content was 1 month in fish of group (i), in which it was probably due to starvation, and 4 to 5 months in group (iii), in which low food supply coincides with the need to draw on body protein to build up the gonads; group (ii) showed a less marked hydration than that in group (iii), but of similar duration. These fish were all from one area; date and duration of max. hydration might be different for other areas. M. D. ANDERSON.

Effect of time and temperature of cooking on the palatability and cooking losses of frozen Atlantic codfish filets. I. L. Armstrong, E. W. Park, B. A. McLaren and D. S. Parker (*J. Fish. Res. Bd. Can.*, 1960, **17**, 1—7).—Commercial frozen codfish filets were defrosted in six ways before cooking at 300 and 500°F respectively. The effects of cooking were assessed by organoleptic, cooking loss and moisture content determinations. Best results were obtained by defrosting by submersion in tap water for 50 min. and cooking, uncovered, at 300 or 500°F. J. V. Russo.

Changes in adenosinetriphosphatase (ATP-ase) activity and sulphhydryl groups of cod flesh during frozen storage. J. J. Connell (*J. Sci. Fd Agric.*, 1960, **11**, 245—249).—Myosin ATP-ase activity of cod muscle is stable at -29° , no decrease in activity being evident on storage up to 3 years. At this stage about half the protein is insol. At -14° and -22° and after storage for 20—25 weeks or 140—180 weeks, respectively, loss of activity was the same, $\sim 55\%$, and the protein was insol. During frozen storage there is no change in either easily-reactable or total sulphhydryl groups. E. M. J.

Variations in chemical composition of different parts of halibut flesh. C. E. Thurston and P. P. MacMaster (*Food Res.*, 1960, **25**, 229—236).—Data indicated that the edible portion of halibut has a higher protein content and much lower oil content than was previously thought. It has a lower Na content than have most salt-water fish and some fresh water fish. Differences between right and left sides were not significant except for oil content. Steak sections were quite uniform in composition viz., protein $\sim 21\%$; oil $< 1\%$ and ~ 50 mg. of Na/100 g. of fish; major variations occurred in the minor parts (dark meat, etc.). A small amount of flesh, not edible, contained $\sim 30\%$ of oil of high quality. E. M. J.

Cystine and total sulphhydryl content of unspoiled and spoiled shrimp. C. H. Kurtzman, D. G. Snyder and H. W. Nilson (*Food Res.*, **25**, 237—244).—There is an increase in total sulphhydryl content of spoiled shrimp. The bacterial population was ~ 500 -fold greater in the spoiled than in the unspoiled shrimp and this may be responsible for the increase in cystine and ammoniacal N of the spoiled shrimp. The possibility of using the increase in cystine content as a chemical index of spoilage is discussed. (31 references.) E. M. J.

Indole and trimethylamine tests for oyster quality. D. Lartigue, A. F. Novak and E. A. Fieger (*Food Technol.*, 1960, **14**, 109—112).—Indole and trimethylamine concn. showed no definite pattern during storage and these tests are not recommended for assessment of oyster quality. Bacterial counts and liquid measurements closely

paralleled organoleptic ratings. Spoilage was evident from the 18th to 20th day of storage, when the bacterial count was $\sim 10^7/g.$ and loss in wt. was $\sim 13\%$. Chlorotetracycline limited bacterial count and development of off-odours. (12 references.) E. M. J.

Spices, Flavours, etc.

Taste interrelationships. R. M. Pangborn (*Food Res.*, 1960, **25**, 245—256).—The taste interactions of sucrose, citric acid, NaCl and caffeine at sub-threshold, threshold and supra-threshold levels, in aq. solutions and more complex mixtures of fruit nectars and tomato juice are described. In general all compounds were found to depress the intensity of each other, e.g., reduction of sweetness of sucrose by citric acid and reduction of sourness of citric acid by sucrose. (25 references.) E. M. J.

Application of the paired comparison method to the study of flavour differences in cooked vegetables. J. Gordon and I. Noble (*Food Res.*, 1960, **25**, 257—262).—Cabbage, cauliflower and broccoli were milder in flavour when cooked in boiling water than when cooked in steam; were similar when cooked by various steaming methods; only broccoli cooked in pressure saucepan was milder than that cooked in steamer and tightly covered saucepan. E. M. J.

Determination of saccharin. P. M. Parikh and S. P. Mukherji (*Analyst*, 1960, **85**, 25—26).—The saccharin is pptd. from a solution acidified with HOAc by known amount of AgNO₃. Residual AgNO₃ is determined in the filtrate by addition of HNO₃, an Fe^{III}NH₄(SO₄)₂ indicator and titration with NH₄SCN at pH < 6 . In presence of Cl⁻ AgCl is redissolved with warm HNO₃. Sodium cyclamate does not interfere. A. O. JONES.

Colouring matters

Colouring matter of blackcurrants. L. Reichel and W. Reichwald (*Naturwissenschaften*, 1960, **47**, 41).—The natural colouring matter was prepared on the lead salt and two-dimensional paper chromatography gave four pigments. Partition chromatography on cellulose columns and development with 0.25% formic acid gave two fractions of which the first was further fractionated on paper. The first fraction contained (a) cyanidin, glucose and rhamnose (β -3-cyanidin-rutinoside) and (b) delphinidin, glucose, rhamnose (β -3-delphinidin-rutinoside). The second fraction contained delphinidin and glucose. Both rutinosides seem to be the pharmacologically active components of blackcurrants. M. LAPIDOT.

Colouring matter of the black elderberry. L. Reichel and W. Reichwald (*Naturwissenschaften*, 1960, **47**, 40—41).— β -3-Cyanidin-primveroside has been synthesised and comparison with the natural pigment sambucyanin proved that the latter is not primveroside, neither is the isoprimveroside which has also been synthesised. Methylation and hydrolysis gave 3,4,6-trimethyl glucose, and the biose component would be 2-(xylosido)- β -D-glucose. Sambubiose is the name proposed for this new sugar, and sambucyanin should be a β -3-cyanidin-sambubioside. M. LAPIDOT.

Preservatives

Non-medical uses of antibiotics. I. Food preservation. R. Levin (*Chem. Prod.*, 1960, **23**, 105—108).—A brief review. C. V.

Preservative effect of antimicrobial agents on high-moisture dried fruits. F. S. Nury, M. W. Miller and J. E. Brekke (*Food Technol.*, 1960, **14**, 113—115).—Processed, high-moisture prunes (35% flesh moisture) without added preservative were submerged for 2 min. in solutions of different concn. of K sorbate (I), Ca propionate, dehydroacetic acid and vitamin K. After draining, they were sprayed with a suspension of a mould or a yeast isolated from spoiled prunes. Other samples were submerged in water for 2 min., drained and inoculated with a mould or yeast and propylene oxide (II) was added to the package before sealing. I and II were the most effective preservatives. E. M. J.

Preservation of fruit juices with Neocytin B (benzyl monobromacetate). A. Schaller and W. Saller (*Fruchtsaft-Industr.*, 1960, **5**, 54—61).—The juices of apples, grapes, plums and tomatoes were prepared with Neocytin B, a recently suggested preservative, and compared with juices preserved by heat treatment. Total and reducing sugars, ash, volatile acids, titratable acids, lactic acid, alcohols, L-ascorbic acid, viscosity and org. Br compounds were determined. References for the methods used and results are given. Added benzyl monobromacetate decomposes after a short storage period. The juices in general had a lighter colour than those preserved by heat, and were rated equal as regards smell and taste after a storage period of > 5 months. (32 references.) I. DICKINSON.

Keeping quality of Pacific coast dogfish. R. H. Moyer, B. A. Southcott, E. G. Baker and H. L. A. Tarr (*J. Fish. Res. Bd. Can.*, 1959, **16**, 791—794).—Experiments aimed at determining the length of time that Pacific coast dogfish can be stored in crushed ice and refrigerated sea water with and without the addition of chlortetracycline are described. Spoilage was assessed by determination of viable bacteria, total volatile base (I) and trimethylamine (II)-N. No appreciable increase in I or II of the muscle was noted until the fish were stored >2 weeks and then increases were small.

J. V. Russo.

Cured vanilla extract from green vanilla beans. McCormick & Co. Inc. (B.P. 812,443, 23.8.56. U.S., 23.8.55).—Cured vanilla extract, in which greater retention of vanilla flavour is attained, is obtained by preparing green bean extract from green vanilla beans (with water) in absence of O₂, concentrating the extract *in vacuo* (after adding an enzyme), then curing the concentrate, viz., by heating at 20—100° during 12—300 hr. while passing therethrough a mixture of inert gas (N₂) and O₂ (0.1—30 vol.-%). After curing, aq. EtOH is added to provide an extract of desired strength, suitable for use as flavouring.

F. R. BASFORD.

Sodium chloride composition. Columbia-Southern Chemical Corp., Asses. of F. Waldo (B.P. 813,164, 15.10.57. U.S., 16.10.56).—NaCl (for table use) is rendered non-agglomerating by compounding with 0.1—5 (0.5—1) wt.-% of finely divided NaAl silicate of particle size <0.1 μ, surface area 10—200 sq. m. per g. Preferably there is employed a silicate analysing as follows: SiO₂ 67, Al₂O₃ 12, Na₂O 9, ignition loss 9%, the balance consisting of Fe, Ca and Ti compounds, and sulphates.

F. R. BASFORD.

Food Processing, Refrigeration

Chemical inactivation of enzymes in vegetables before dehydration. R. U. Makower (*Food Technol.*, 1960, **14**, 160—164).—The tissues were infiltrated under a vac. with various chemicals and the degree of inactivation of catalase, peroxidase, acetyl esterase, phosphatase and polyphenol oxidase were determined. Acids, ethanol and surfactants were used alone and in combination. Nearly complete inactivation was achieved in cabbage, celery, carrots and apples. In general the anionic surface-active compounds were the most effective synergistic enzyme inhibitors in the acid-ethanol mixture, cationic compounds were least effective. Peroxidase seemed to be the most difficult to inactivate. (21 references.)

E. M. J.

Effect of heat and irradiation on the microflora of canned hams. S. D. Drake, J. B. Evans and C. F. Niven, jun. (*Food Res.*, 1960, **25**, 270—278).—*Streptococcus faecium* is the most troublesome bacterial organism to resist normal commercial heat processing or irradiation with approx. 1 megarad of γ-irradiation. Except for these spores and those of bacilli and clostridia most of the flora (lactic acid bacteria) are destroyed. Heat and irradiation offer no special advantage over present methods of heat processing and frozen storage.

E. M. J.

Inactivation of type A *Clostridium botulinum* toxin by irradiation with cobalt-60. R. O. Wageraar and G. M. Dack (*Food Res.*, 1960, 279—284).—Purified type A botulinum toxin (I) in Na phosphate buffer was readily inactivated by irradiation. I was added in the same concn. to 0.05M Na phosphate buffer solution (pH 7.5), sterile 5% trypticase broth and heat-treated surface ripened cheese. These three prep. were subjected to γ-irradiation from ⁶⁰Co at several dosage levels. The addition of 5% trypticase broth to I increased the irradiation dosage necessary to inactivate I ~100-fold compared with irradiation of toxin in Na phosphate buffer solution. I was protected from inactivation slightly more by the cheese medium than by broth.

E. M. J.

Resistance of *Bacillus coagulans* spores to γ-rays. Application of the multiple tube probability method. A. Anellis, C. J. Chichon and M. M. Rayman (*Food Res.*, 1960, **25**, 285—295).—The method is described. Factors affecting the radiosensitivity of *B. coagulans* spores were: gaseous environment, protective nature of the medium and initial concn. of the spores. D₉₀ (dose required to eliminate 90% of contaminant organisms) values were 24% greater when phosphate buffer suspensions were irradiated in presence of N₂ instead of air. The resistance in protein-fortified skim milk medium was 43% higher than in phosphate buffer when the gaseous environment for each substrate was N₂. (21 references.)

E. M. J.

Microbial spoilage of canned food. II. Effect of heat, H-ion concentration, and chemicals on spoilage bacteria. G. Rangaswami and R. Venkatesan (*Proc. Indian Acad. Sci.*, 1960, **51B**, 9—18).—Bacteria isolated from spoiled canned foods were examined for their sensitivity to heat treatment, pH, and chemical preservatives.

Spp. of *Bacillus* (*B. circulans*, *B. brevis*, *B. subtilis*, *B. coagulans* and *B. licheniformis*) were highly resistant to heat, *Clostridium histolyticum* was less resistant, and *Lactobacillus fermenti* was instantly killed at 100°. All spp. grew over a wide range of pH, *L. fermenti* even at pH 3.0. *L. fermenti* and *Cl. histolyticum* were sensitive to low concn. of salt, but *B. licheniformis* tolerated 13% of salt. All spp. were inhibited by sugar at a concn. of 40 to 45° Brix; *Cl. histolyticum* was inhibited at 30° Brix. Na benzoate and K metabisulphite were synergistic in their action on bacteria, and were more effective at acid or alkaline pH than at neutral pH. Both substances were synergistic with sugar. (19 references.)

M. D. ANDERSON.

Time-temperature tolerance of frozen foods. XX. Boysenberries. D. G. Guadagni, K. M. Eremia, S. H. Kelly and J. Harris (*Food Technol.*, 1960, **14**, 148—150).—Effect of temp. in larger than retail packages of frozen fruits, e.g., individually quick frozen (IQF) form and packaged in institutional sizes for remanufacture into pies, was studied. Data are given on moisture, ascorbic acid contents, colour and flavour changes in IQF and syrup-packed frozen boysenberries held at temp. 0—30°F.

E. M. J.

Fish freezing. J. W. Slavin (*Industr. Refrig.*, 1959, **137**, No. 3, 18—19, 22—24).—Methods and equipment in commercial use are reviewed. (29 references.)

C. V.

Preservation of food and other perishable materials. British Oxygen Co. Ltd. (812,210, 21.11.56).—A method of preserving food or other perishable material during transport or storage comprises liquefying air or N₂ with liquid methane as refrigerant and cooling the material therewith in a thermally insulated container below -50 (-190)°. Apparatus is figured and claimed.

F. R. BASFORD.

Packaging

Collapsible polyethylene tube as a food package. J. D. Kemp, R. M. Ballantyne, A. J. Duckes and J. W. Haynes (*Food Technol.*, 1960, **14**, 131—134).—Jam, jelly, honey, groundnut butter and creamery butter samples were stored at 0, 70 and 100°F in lined and unlined tubes. Tubes lined with polyvinylidene chloride resin were more suitable because of less tendency to produce off flavours and impermeability to oils; O₂ transmission characteristics need further study. At 100°F, the storage life of jams, jelly and honey in lined tubes was generally comparable to that in cans or bottles for periods up to >3 months.

E. M. J.

Miscellaneous

Nutrition, proteins, amino-acids, vitamins

Nutritive value of protein foods based on blends of groundnut, soya-bean, Bengal gram (*Cicer arietinum*) and sesame flours. K. Krishnamurthy, S. N. Ganapathy, R. Rajagopalan, M. Swaminathan and V. Subrahmanyam (*Food Sci., Mysore*, 1959, **8**, 388—389).—Weanling rats received as sole source of protein either dried skim milk (I), or a protein food containing low-fat groundnut, soya and Bengal gram flours (II), or the same with low-fat sesame flour replacing some of the groundnut flour (III). Protein efficiency ratios in 8-week tests were I 2.55, II 1.70, III 2.13. With 16% of protein in the diet, the average weekly gains of wt. were the same on all 3 sources of protein.

M. D. ANDERSON.

Chemical composition and nutritive value of Bengal gram (*Cicer arietinum*). M. N. Rao, R. Rajagopalan, M. Swaminathan and V. Subrahmanyam (*Food Sci., Mysore*, 1959, **8**, 391—395).—Bengal gram flour contains 12 to 31% of protein, the lowest values being due to absence of nodule bacteria. Globulin accounts for 88% of the total N. The proteins are good sources of all essential amino-acids except tryptophan and methionine. They contain more lysine and threonine than groundnut proteins, and therefore supplement cereal proteins more effectively. Bengal gram is a fairly rich source of B vitamins. The biological values of Bengal gram protein for rats when fed at 5, 10 and 15% of the diet were 60, 52 and 46, and the digestibilities 85, 88 and 69. The protein efficiency ratio at 10% has been reported as 1.27 and 1.48. A diet of Bengal gram + cod-liver oil and mineral salts permitted growth and reproduction in rats, but not lactation. Bengal gram could be used with groundnut flour for the prep. of Indian multi-purpose food. (40 references.)

M. D. ANDERSON.

Proximate composition of some commercial Indian biscuits. K. M. Narayanan, N. S. Kapur and D. S. Bhatia (*Food Sci., Mysore*, 1959, **8**, 387—388).—Figures are given for the pH, and the contents

of water, crude protein, fat, ash, reducing sugar, invert sugar, sucrose and carbohydrate, in 18 samples of Indian biscuits, 15 sweet and 3 salt.
M. D. ANDERSON.

Comparison of the amino-acid composition of protein in flour and endosperm from different types of wheat, with particular reference to variation in lysine content. E. E. McDermott and J. Pace (*J. Sci. Fd Agric.*, 1960, **11**, 109—115).—Flours from Manitoba and Hybrid 46 wheats differing greatly in protein content and in the physical characters exhibited by the protein resembled each other in amino-acid composition. There were small differences with respect to the content of lysine, arginine and cystine. Examination of six other flours from different wheats differing in protein content and physical characteristics showed that lysine content varies inversely with the protein content. Samples of endosperm with a vitreous character compared with those of a mealy type from grains of the same sample of the same variety of wheat had a higher content of protein, but the lysine and arginine content of the protein was lower. (10 references.)
E. M. J.

Assay of proteolysed food proteins and their products. G. C. Esh, S. K. Ganguly and U. P. Basu (*J. Instn Chem. India*, 1959, **31**, 160—166).—For proper nutritional assessment, the estimation of (a) total N, (b) NH_3 , (c) amino-N, (d) amino-acid composition and (e) biological adequacy is discussed with particular reference to meat hydrolysates from papain and pancreas enzymes. An improved method for (b), by distilling the NH_3 with a phosphate buffer at pH 7, gave nearly theoretical results. Satisfactory results were also obtained for (c) using pure amino-acids and protein hydrolysates by a modified formol titration (pH of hydrolysate, formaldehyde and final titration adjusted to 6.0, 7.5 and 9.0 respectively). The rat depletion-repletion method was used for assaying (e) using 4—5% of various hydrolysates on a 12 day test.
P. M. KINGSTON.

Use of detergents for removal of nitrogen from plant materials. A. Bevenue and K. T. Williams (*J. Ass. off. agric. Chem., Wash.*, 1959, **42**, 441—444).—1% aq. solutions of Santomere No. 1 (an alkylarylsulphonate) and Duponol C (sodium dodecyl sulphate) containing 0.5% of Na_2CO_3 removed 92—95% of the protein N from lima bean, navy bean and split pea meals. The latter detergent removed only a trace of the hemicellulose component of lima beans. All of the non-ionic surface active agents tested in 1% and 2% aq. solution were less efficient than Na_2CO_3 for removing N.
A. A. ELDRIDGE.

Nutritional-physiological effects of present method of manufacture of milk powder: can adverse effects be eliminated by subsequent Yoghurt fermentation? II. H. Fink, U. Ruge and I. Benda (*Fette Seif. Anstrichm.*, 1960, **62**, 292—294).—The incidence of liver necrosis in growing albino rats fed on a diet containing skimmed milk powder may be reduced but not eliminated by curd fermentation of the regenerated milk.
J. L. PROSSER.

Colorimetric determination of amino-acid concentration. W. A. Vincent (*Nature, Lond.*, 1960, **185**, 530).—The sample solution is incubated for ~12 hr. at pH 7 with a 1% aq. suspension of $\text{Cu}_2(\text{PO}_4)_2$ and, after centrifugation, the concn. of Cu in the supernatant is determined spectrophotometrically at 440 μ with Na diethyldithiocarbamate. The relation $[\text{Cu}]/[\text{amino-acid}]$ is linear over a wide range, so that dilution is permissible. Accuracy is within $\pm 2\%$ for 50—1000 $\mu\text{g}/\text{ml}$. (α -alanine). Peptides and proteins do not interfere.
W. J. BAKER.

Direct amino-acid analysis by gas chromatography. A. Zlatkis, J. F. Oro and A. P. Kimball (*Analyt. Chem.*, 1960, **32**, 162—164).—The sample is injected into a flowing system whereby a heated microreactor containing ninhydrin oxidises the amino-acids to aldehydes + CO_2 which are separated in a chromatographic column. After leaving the column the aldehydes are cracked to CH_4 and water, water is removed and CH_4 is measured by a thermal-conductivity cell.
G. P. COOK.

Water-soluble, ninhydrin-positive substances retained by anion-exchange resin. F. Lindlar (*Naturwissenschaften*, 1960, **47**, 14).—Retention of water-sol. ninhydrin-positive substances by Amberlite IR-45 in chromatographic separation of trichloroacetic acid from amino-acid mixtures was investigated by high-tension paper electrophoresis using pyridine-glacial acetic acid-water as buffer and n-heptane as solvent. The dried spherogram, developed with ninhydrin, showed that relative composition of the mixtures did not vary, whether elution was carried out at room temp. or at the boil with 6N-HCl.
W. H. KEMP.

Determination of monomers during polycondensation of α -amino-acid esters by hydrochloride method. T. D. Kozarenko, N. B. Noskova and K. T. Poroshin (*Izv. Akad. Nauk SSSR, Otd. khim. Nauk*, 1959, 1324—1327).—Speed of reaction of the monomers during the first stage of polycondensation of α -amino-acid esters

was examined and reaction mechanism studied during formation of peptide bonds in presence of CO_2 . The difference between initial and final amounts of monomer was assessed in the usual way, separation of unreacted monomers being by extraction with alkyl sulphate. An improved method for determining monomers in the alkyl sulphate was by ppting them as hydrochlorides (as white crystals or oil). The ppt. separate quant. in vac. at moderate temp.; they are not affected by heating to 100° and are capable of further polycondensation. The method can be applied for esters of DL-alanine, DL-valine, L-proline and DL-phenylalanine. Loss of the hydrochlorides is $> 1.5\%$.
A. L. B.

Responses of two strains of rats to rapeseed oil and maize oil. J. L. Beara, T. K. Murray and J. A. Campbell (*Canad. J. Biochem. Physiol.*, 1960, **38**, 187—192).—On a basal diet of ground fox cubes, with 20% of rapeseed oil or maize oil, Sprague-Dawley and Wistar rats differed in the following respects. (i) The effect of rapeseed oil in lessening rate of gain of wt. was much more marked in Wistar rats. (ii) Rapeseed oil and maize oil had similar apparent coeff. of digestibility in Wistar rats, but in Sprague rats the rapeseed oil had a lower coeff. of digestibility. (iii) After 6 weeks on the diets, the testes of Wistar but not of Sprague rats were smaller on the rapeseed than on the maize oil, and (iv) the adrenal glands of Sprague but not of Wistar rats were larger on the rapeseed oil. These differences between strains of rat explain some discrepancies in the reported effects of rapeseed oil on rats. Strain of rat and type of oil had no effect on the amount of vitamin A stored by the liver, or on the rate of depletion of vitamin A from the liver when a single dose of vitamin A was given after a period of vitamin A deficiency. (12 references.)
M. D. ANDERSON.

Activated [Yugoslav] hydrosilicate earth in determination of nicotinic acid. M. Filajdić and V. Mikulić (*Kem. u Industr. Zagreb*, 1960, **9**, 1—3).—A type of Yugoslav activated earth (BVR) from Kutina is a good substitute for Frankonite, originally recommended for the La Roche method (I) of nicotinic acid determinations. Results obtained for wheat by I were somewhat higher than those by the microbiological method.
A. L. GROCHOWSKI.

Partition column for vitamin-A chromatography. J. B. Wilkie, S. W. Jones and W. W. Morris (*J. Ass. off. agric. Chem., Wash.*, 1959, **42**, 422—423).—The partition system employing a polyethylene glycol 600 as the immobile phase with Celite 545 as support and iso-octane as the mobile phase (Theivagt and Campbell, *Analyt. Chem.*, "in press") is improved by the use of light petroleum, b.p. 30—60°, as a supplement in the mobile phase. With this procedure acceptable results were obtained for recovery and reproducibility in the determination of vitamin A.
A. A. ELDRIDGE.

Radiosynthesis of vitamins C and B₁ in foods. G. M. Egiazarov (*Dokl. Akad. Nauk SSSR*, 1959, **128**, 1070—1072).—The effect of small γ -irradiation doses (25,000—150,000 r., ^{60}Co source) on the vitamin contents of a large no. of foods was investigated. Irradiation was carried out in dark at 5—8° at 18,000 r./hr. In general the vitamin contents were reduced proportionally to the irradiation doses, but at 50,000 r. doses the vitamin C concn. in potatoes and carrots, and the vitamin B₁ contents in pearl barley and buckwheat cereal were increased. Larger radiation doses decreased the vitamin contents. In potatoes and carrots the additional synthesis of ascorbic and dehydroascorbic acids on small γ -irradiation was considered in connexion with the defensive functions of the vegetable tissue and the known healing processes. In processed foods such as tomato paste and sauerkraut, and in synthetic vitamin concentrates, no such increase took place. In wheat and rye flour and in millet the same radiation doses resulted in a considerable decrease in the thiamine contents.
A. L. GROCHOWSKI.

Unclassified

Problems in the preparation and handling of hot vended canned foods. G. T. Peterson, J. F. Fox and L. E. Martin (*Food Technol.*, 1960, **14**, 89—91).—The use of coin-operated machines equipped with facilities to heat specially prepared canned foods to serving temp. (140—150°f) is discussed. Typical products include soups, meat stews, spaghetti with meat balls, chicken noodle dinner, etc., in cans of capacity 6—8½ oz., etc. The chief problems are concerned with: bacteriological aspects of prep., storage and handling of the hot canned goods, effect of high temp. storage in the machines on product quality and consumer acceptance of the goods.
E. M. J.

Composition of South Pacific foods. F. E. Peters, M. Tomono and P. A. Wills (*Food Res.*, 1960, **25**, 211—228).—A nutritional survey is presented covering data on proximate composition of 62 local foods, the amino-acid composition of two staple foods (taros and yams) and on the vitamin content of 38 foods used. (24 references.)
E. M. J.

Identification of pesticide residues in extracts of fruits, vegetables and animal fats. I. Chromatography. W. P. McKinley and J. H. Mahon (*J. Ass. off. agric. Chem., Wash.*, 1959, **42**, 725—734).—Procedures are described for the extraction of pesticide residues with benzene or CCl_4 , purification of the extracts by partition between n-hexane and acetonitrile, adsorption of residual fat on a Florisil column and for paper chromatography on Whatman No. 1 paper with three solvent systems, (a) immobile phase, 2-phenoxyethanol (14% in ether); mobile phase, 2,2,4-trimethylpentane, (b) immobile phase, liquid paraffin B.P. (4% in ether); mobile phase, 40% aq. pyridine, and (c) immobile phase as for (b); mobile phase 70% aq. acetone. R_f values (referred to lindane as reference standard) are tabulated for 52 compounds. Variations of technique suitable for particular problems are discussed. In a study of >1000 samples of fruit and vegetables, 80% of samples contained <1 p.p.m. of DDT and its degradation product DDE, and 3% contained more than the tolerance level of 7 p.p.m. of DDT. (13 references.)

F. C. APLING.

Detection and semi-quantitative estimation of chlorinated organic pesticide residues in foods by paper-chromatography. P. A. Mills (*J. Ass. off. agric. Chem., Wash.*, 1959, **42**, 734—740).—Suitable extraction procedures are detailed for pesticide residues in large fruits; small soft fruits, vegetables, beans and dry feeds; fats and oils; cheese; milk; and animal tissues. Purification of the extracts by acetonitrile-hydrocarbon partition and by columns of Florisil, MgO -Celite and H_2SO_4 -Celite is described. For the paper chromatography on Whatman No. 1 paper, suitable solvent systems are (i) aq. immobile solvent, heavy mineral oil USP (or maize, cottonseed, tung or soya oil), 5% in ether; mobile solvent, 75% aq. acetone, Methylcellosolve or methanol, or 40% aq. pyridine, and (ii) non-aq. immobile solvent, NN' -dimethylformamide, 2-phenoxyethanol or dimethylcyanamide, 35% in ether; mobile solvent, 2,2,4-trimethylpentane or mixed octanes. For location of spots and semi-quant. estimation the dried papers are sprayed with a reagent containing AgNO_3 , water, 2-phenoxyethanol, 30% H_2O_2 and acetone. The sprayed papers can then be exposed to u.v. light until the reduced Ag spots are developed. Recovery data are reported for 13 pesticides, and vary from 40 to 100%. (13 references.)

E. C. APLING.

Detection and identification of antimicrobials by paper chromatography. C. Genest and R. A. Chapman (*J. Ass. off. agric. Chem., Wash.*, 1959, **42**, 436—440).—Antimicrobial substances likely to be found in foods may be separated and identified by paper chromatography. In system I the immobile solvent is formamide in acetone, the mobile solvent is acetic acid in CHCl_3 , and the chromogenic agents are KMnO_4 in aq. Na_2CO_3 , FeCl_3 , and AgNO_3 in aq. ammoniacal alcohol; *p*-hydroxybenzoic acid, salicylic acid, benzoic acid, sorbic acid and dehydroacetic acid are detected. In system II the immobile solvent is water, the mobile solvent is ammoniacal aq. acetone and the chromogenic agents BDH Universal Indicator; phthalic acid and aniline in water and n-butanol; PtCl_4 and KI; sodium diacetate, propionic acid, hexamethylenetetramine and Na_2SO_3 are detected. In system III the immobile solvent is formamide in acetone, the mobile solvent is NET_3 in cumene, and Millon's reagent or illumination by u.v. light is employed; methyl, ethyl, propyl, butyl and benzyl *p*-hydroxybenzoate are thus detected.

A. A. ELDRIDGE.

Food engineering for atomic submarines. C. M. Schoman, jun. (*Food Technol.*, 1959, **13**, 615—620).—Facilities for improving food service are described.

E. M. J.

Survey of radioactive residues in foods before and after 1945: evidence of possible fallout contamination. E. P. Laug and W. C. Wallace (*J. Ass. off. agric. Chem., Wash.*, 1959, **42**, 431—436).—The foods were ashed and the beta-radioactivity was measured with an instrument having 100% response to β -particles but a low response to γ -photons. ^{40}K was assumed to be uniformly distributed in nature; its γ -component was ignored. KCl (100 mg.) was used as the working standard. The ash samples were analysed for K with a flame photometer, and the contribution of the ^{40}K was subtracted from the total radioactivity to give the net radioactivity. Errors are discussed and results tabulated. Fish, shellfish, dairy products and tea (particularly Japanese, Formosan, Malayan and Indian) showed significant increase of radioactivity if sampled after 1945.

A. A. ELDRIDGE.

3.—SANITATION

Animal fragments in cereal products. G. B. Wagner (*Cereal Sci.*, 1960, **5**, 63—64, 75).—Insect, rodent and bird contaminants in human cereal foods can come from the raw grains, the flour mill, the air, or the surface of sacks, machinery or packets. If the fragments

are identifiable it is often possible to trace the source of contamination. Further efforts are needed to prevent this contamination. S. G. AYERST.

Influence of oil content on the susceptibility of seeds to fumigation with methyl bromide. R. E. Blackith and O. F. Lubatti (*J. Sci. Fd Agric.*, 1960, **11**, 253—258).—The susceptibility of several types of oily seed is examined over a range of moisture contents and dosages. The presence of oil in seeds, of itself, renders them only more slightly susceptible to MeBr , but the availability of water present in the seed is increased. The deleterious influence of a high moisture content is enhanced in oily seeds, in respect of fumigation damage and deterioration in storage. The oil may act as a reservoir for the fumigant and germination may be delayed up to a month in some seeds. Because of the wide variation in oil contents of seeds the oil- as well as the water content of the seed should be determined before fumigation. E. M. J.

Penetration of piperonyl butoxide as a synergist and as an antagonist in *Musca domestica*, L. G. W. Ware (*J. econ. Ent.*, 1960, **53**, 14—16).—Piperonyl butoxide disappeared more rapidly in first 24 hr. when applied alone than when applied with either malathion or sevin. There is a higher percentage penetration at 1.25 $\mu\text{g}/\mu\text{l}$. than at 3.5 $\mu\text{g}/\mu\text{l}$. C. M. HARDWICK.

Syntheses of the aliphatic deuterium analogues of DDT and TDE and their toxicity and degradation when applied to adult houseflies. R. J. Barker (*J. econ. Ent.*, 1960, **53**, 35—41).—Absorption spectra showed that the deuterium analogues had the same electron pattern. Female flies absorbed more DDT and d-DDT than males. The process of degradation in the housefly and in alkali is compared. (28 references.) C. M. HARDWICK.

Cumulative effect of sublethal doses of myristyl chloroacetate on a highly DDT-resistant strain of *Musca domestica*, L. S. Bettini, M. Boccacci and G. Natalizi (*J. econ. Ent.*, 1960, **53**, 99—101).—Three-day-old female flies were exposed to sub-lethal doses with different time intervals. With intervals up to 120 hr. accumulation of toxic effects occurred. (12 references.) C. M. HARDWICK.

Metabolism of Co-Ral (Bayer 21/199) by tissues of the house fly, cattle grub, ox, rat and mouse. R. D. O'Brien and L. S. Wolfe (*J. econ. Ent.*, 1959, **52**, 692—695).—Co-ral is metabolised only in the liver in mammals. The mouse activates it and is therefore susceptible; the ox and rat degrade it and are resistant. Both activation and degradation are found in the cow. Whole fly minces readily activated Co-ral but only after the use of four techniques was activation found in *Hypoderma bovis*. It was located in the gut and fat body. The effectiveness of dermal over oral application against cattle grubs can be explained by the fact that Co-ral will reach the grubs before it reaches the liver. C. M. HARDWICK.

Insecticidal activity of enantiomorphs of O-ethyl S-2-(ethylthio)-ethyl phosphorothiolate. T. R. Fukuto and R. L. Metcalf (*J. econ. Ent.*, 1959, **52**, 739—740).—The *l*-isomer inactivates fly brain cholinesterase 11 times faster than the *d*-isomer and 1.8 times faster than the *d-l*-mixture. The *l*-isomer was 6, 10 and 9.7 times more toxic by contact to the house fly, mosquito larvae and honey bee, respectively, than was the *d*-isomer. C. M. HARDWICK.

Toxicological action of DDT on three species of mosquito larvae. D. E. Weidhaas and C. H. Schmidt (*J. econ. Ent.*, 1960, **53**, 106—110).—Radiometric techniques showed that 3 spp. of 4th instar larvae absorbed more DDT at any given concn. with increased time or in any given time with increased concn. Mortality was not directly related to the dose absorbed; length of exposure and concn. were also significant. Differences in mortality between species were not related to the amount absorbed. Excretion of DDT occurred only after exposure to a high concn. for a short period. Live larvae absorbed more than dead ones. C. M. HARDWICK.

Effects of heterogeneous distribution and codistillation on the results of tests with DDT against mosquito larvae. D. E. Weidhaas, C. H. Schmidt and M. C. Bowman (*J. econ. Ent.*, 1960, **53**, 121—125).—The amount of DDT in suspension in different sized containers both open and sealed was estimated radiometrically and biologically. Codistillation and association with the water interface affected the mortality data for *Anopheles quadrimaculatus* more than that for *Aedes aegypti*. Parathion, malathion, lindane and dieldrin were not affected sufficiently to alter mortality. C. M. HARDWICK.

Control of the oriental rat flea with systemic insecticides fed to rats. T. L. Harvey (*J. econ. Ent.*, 1960, **51**, 167—168).—Oral treatment of rats with Ronnel, Dowco 109 or Dimethoate gave significant kills of *Xenopsylla cheopis* feeding on them in 3 days. When 400 fleas were exposed to two Dimethoate-treated rats, 80% kills were obtained in 2 days. C. M. HARDWICK.

Systemic insecticide and bait for flea and rat control. E. Bennington (*J. econ. Ent.*, 1960, **53**, 169—170).—If given with alternatives, Ronnel-containing bait with or without fumarin was avoided by the rats. Baits containing 2.4 g./lb. were taken well by laboratory and wild rats but 6-g. and 12-g. baits were often left. The addition of sugar and a commercial liquid-smoke condiment to 12-g. baits killed all fleas before the rats died, after 5 days feeding. Baits remained free of insects. C. M. HARDWICK.

Aerosol disinfection of poultry premises. D. A. McKenzie, J. Lambert and J. Getty (*J. appl. Bact.*, 1959, **22**, 258—263).—A proprietary trichloroethylene glycol mixture was examined to ascertain the effect on atm. and surface disinfection. Only moderate success was attained using *Bacterium coli* as test organisms; an increase in concn. did not improve on 75% kill which was attained after 30 min. exposure. With formalin-water aerosol, very satisfactory results were attained. C. V.

Toxicity of malathion to Killifish (Cyprinodontidae) in Delaware R. F. Darsie, jun., and F. E. Corriden (*J. econ. Ent.*, 1959, **52**, 696—700).—Malathion was sprayed at the rate of 2 lb./acre on to tubs containing *Fundulus ocellaris*. After 4 hr. 26% were dead and 42% showed sub-lethal poisoning and of these 56% had recovered after 64 hr. in fresh water, and only 8% remained moribund. There was no correlation between the size of fish and mortality. (15 references.) C. M. HARDWICK.

Coli-aerogenes bacteria in farm water supplies. S. B. Thomas, P. M. Hobson and R. G. Druce (*J. appl. Bact.*, 1959, **22**, 32—45).—In this group 825 cultures were isolated at 30° and 735 at 37° from 645 samples. *Klebsiella* (I) were present in 50% of those isolated at 30° and *Escherichia coli* (II) were the dominant type, 57%, in the 37° group. *Citrobacter freundii* (III) and *K. cloacae* were found in waters of high sanitary quality derived from springs and wells which were protected. Fortnightly sampling of 11 supplies showed marked seasonal variation; II were high in summer while 37°-negative strains of I and III were higher in winter. (28 references.) C. V.

Dissolved oxygen requirements of cold water fishes. R. C. Davison, W. P. Breeze, C. E. Warren and P. Doudoroff (*Sewage industr. Wastes*, 1959, **31**, 950—966).—The influences of temp. and O₂ supply on fasting juvenile coho salmon, *Oncorhynchus kisutch*, food consumption and growth, and the influence of sulphite-process pulp mill waste (I) on survival were studied. Further observations were made on the resistance of the sculpin *Cottus perplexus* to low O₂ concn. at summer temp. (18—19°). At constant temp. of 20° and less, in summer and autumn, the fasting young salmon tolerated for 1 day O₂ concn. <2 mg./l., but not 1 mg./l. Yearling salmon lived for 30 days at temp. near 18° and O₂ concn. of 2 mg./l., but ate little food and lost wt. At O₂ concn. of 2.9 mg./l. the fish fed well and gained wt. The presence of I (1.2 mg./l.) had no effect on the min. dissolved O₂ requirements of juvenile salmon. Resistance of sculpin to reduced O₂ concn. at 18—19° was similar to that of the juvenile salmon. (17 references.) O. M. WHITTON.

Insecticide pollution of water resources. H. P. Nicholson (*J. Amer. Wat. Wks. Ass.*, 1959, **51**, 981—986).—A review. (13 references.) O. M. WHITTON.

Determination of slimes in rivers. J. N. Wilson, R. A. Wagner, G. L. Toombs and A. E. Becher, jun. (*J. Wat. Pollut. Control Fed.*, 1960, **32**, 83—89).—A trawl to locate active *Sphaerotilus* growth, the tile-box sampler to correlate areas of growth and growth characteristics with concn. of nutrients and physical factors, and the Reighard tow net to determine quality, quantity and distribution of detached and suspended slimes are described. O. M. WHITTON.

Chlorinated insecticides in surface waters. A. A. Rosen and F. M. Middleton (*Analyt. Chem.*, 1959, **31**, 1729—1732).—Small concn. of chlorinated insecticides are analysed by adsorption on an activated C column 18 in. × 3 in. This column is suitable for 5000 gal. of H₂O. The C is extracted with CHCl₃ and the extract concentrated; 0.25—0.5 g. of solid concentrate in 3 ml. CHCl₃ are passed over an alumina column (80—200 mesh) 14 cm. × 1.9 cm. and eluted with CHCl₃ until three column vol. have been collected. The eluted fractions are concentrated on a steam bath and finally *in vacuo* at 50°. The residue is dissolved in 0.2 ml. CS₂ and the i.r. spectrum determined. The method is mostly of value for the qual. detection of insecticides and the following were investigated: aldrin, BHC, chlordane, DDD, DDT, dieldrin, endrin and methoxychlor. (17 references.) S. BAAR.

Cooling of condenser-water. H. Tonn (*Msch. Brauerei wissen. Beil.*, 1959, **12**, 179—187).—A review covering practical and theoretical considerations involved in the economic utilisation of cooling-water and of air-cooling of the used water. P. S. ARUP.

Meat packing plant effluent as irrigation medium. H. A. Voll-

brecht (*Dissert. Abstr.*, 1959, **20**, 824).—In an 8-year experiment on three different soils, irrigation with effluent from a meat-packing plant added N, P, K and Na to the soils. Only small amounts of N, P and K were found in the percolate. About half the N of the effluent was removed by crops. Some was apparently lost by denitrification. Large amounts of soil Ca and Mg were displaced by effluent Na, and were leached from the soil mainly as chlorides and bicarbonates. Hay yields were 100 to 140% greater with irrigation by effluent than with water irrigation, or no irrigation. Water irrigation caused considerable leaching of N. Experiments on the structure of irrigated soils indicated that wastes containing over 1000 p.p.m. of Na would be unsuitable for long-term irrigation of fine-textured soils. There was, however, no structural deterioration when 57 p.p.m. of Ca and 36 p.p.m. of Mg were added to the irrigating fluid as well as 1000 p.p.m. of Na. M. D. ANDERSON.

Wastes from processing of maize chips. R. Porges (*J. Wat. Pollut. Control Fed.*, 1960, **32**, 182—185).—Data from a waste survey of a maize-chip plant were analysed and correlated with production figures and processing methods. O. M. WHITTON.

Rate of oxidation of organic wastes in soil. J. M. Costopoulos (*Dissert. Abstr.*, 1959, **20**, 2194—2195).—Rate of oxidation of an artificial waste (prepared from skimmed milk) in fine sand was studied. Curves of O₂ utilisation were similar to B.O.D. curves in water, showing an induction period and first- and second-stage oxidations. First-stage oxidation was postulated as a first-order reaction; ratio of ultimate B.O.D. to applied C.O.D. was 0.65—0.7, and substantially independent of org. loading. Dependence of available B.O.D. on rate of org. loading, interval of loading, rate of oxidation, and time, is postulated in a theory upheld by experimental evidence. A. M. SPRATT.

Action of algae on sewage in ponds. H. T. Clausen (*J. Inst. Sew. Purif.*, 1959, 345—351).—Observations are reported of the self-purification of settled sewage (I) by algal action in effluent ponds. Figures for B.O.D. reduction with time are obtained: for some mixes of algal effluent with I 60% reduction in 2 hr. was obtained. *Chlorella* predominate during early growth but *Spirulina* are more numerous later. O. M. WHITTON.

Significance of organic nutrition of sewage-grown algae in relation to algal growth in sewage-stabilisation ponds. W. O'F. Pipes, jun. (*Dissert. Abstr.*, 1959, **20**, 2217).—*Chlorella pyrenoidosa* employed in the tests can metabolise some of the dissolved org. material of sewage in absence of bacteria. Growth conditions on inorg. media and in a sewage-stabilisation pond are compared; in the former light is the limiting growth factor, and in the latter it may be either light or CO₂ concn. Org. nutrition of algae contributes to greater yields of algae compared with inorg. cultures. A. M. SPRATT.

Aquatic fungi in water with high waste loads. W. B. Cooke and A. F. Bartsch (*Sewage industr. Wastes*, 1959, **31**, 1316—1322).—Tests show that aquatic fungus population of a stream is not a suitable parameter of water quality. O. M. WHITTON.

Oxidised nitrogen in waters and sewage effluents observed by ultra-violet spectrophotometry. R. C. Heather and R. F. Rackham (*Analyt.*, 1959, **31**, 548—551).—NO₂⁻ and NO₃⁻ (present only in traces) are measured from their absorbance at 210 mμ. The extinction of org. matter at 210 mμ is four times that at 275 mμ, at which wavelength the extinction of NO₂⁻ is negligible. Results with water of moderate org. purity agree with those of other methods. With some samples containing NO₃⁻ freshly formed by microbial action discrepancies suggest the presence of unstable compounds that change slowly to nitrate. The method is suggested as an additional means of investigating nitrification in soils, waters and sewage effluents. A. O. JONES.

Zoogloeal-forming organism in activated sludge. T. C. Buck and C. E. Keefer (*Sewage industr. Wastes*, 1959, **31**, 1267—1274).—A zoogloeal-forming organism was isolated from activated sludge. It was obtained in pure culture and its morphological, biochemical and physiological characteristics were studied. It differed in many ways from *Zoogloea ramigera*. It reduced nitrates with the production of NH₃ and N₂ gas. An Alcian blue stain indicated the presence of polysaccharides in the gelatinous matrix surrounding the cells of the organism. When settled sludge was inoculated with the organism, aerated for 4 hr., and settled for ½ hr., the reduction in B.O.D. was comparable with that obtained when activated sludge was used. O. M. WHITTON.

Utilisation of liquid [sewage] sludge. R. C. Merz (*Wat. & Sewage Wks*, 1959, **106**, 489—493).—This describes the use of liquid, digested sludge to reclaim waste land for agricultural purposes. Sludge loadings of 100 tons of dry solids/acre leave crop growth unimpaired, of 25 tons produce crop growths comparable to those achieved with commercial fertilisers, and of 50 tons produced sufficient fertilisation for a second superior crop. O. M. WHITTON.

Experiments in aeration. S. G. Burgess and L. B. Wood (*J. Inst. Sew. Purif.*, 1959, 258—270).—Investigations were made to compare the efficiency of aeration by aluminum tile diffusers: 7 in. aluminum dome diffusers (I), and 4 in. aluminum dome diffusers; to compare the efficiencies of different I and the effect of spacing on efficiency of I and effects of synthetic detergents on rate of reaeration of water. Data are given on each section of the work.

O. M. WHITTON.

The air-conditioning industry. R. Larose (*Industr. Aliment. agric.*, 1959, 76, 595—600).—Air-conditioning maintains the air within a building at desirable levels of temp. and humidity, renews it, cleans it, and keeps it in movement. The physiological and physical principles involved are outlined. The advantages of air-conditioning for people working in the building, and for the work carried out, are stressed.

M. D. ANDERSON.

Bactericidal compositions. Cadum Palmolive (B.P. 804,450, 5.9.56. Fr., 15.9.55).—A synergistic bactericidal composition, especially useful in the cleaning of equipment employed in the dairy industry, consists of chloramine T (I) and 1—5 wt.-% of a compound R·A·R (R is phenyl group substituted in the *o*-position by OH and elsewhere by at least 1 Cl; A is CH₂ or S), e.g., 5,5'-dichloro- or 3,5,6,3',5',6'-hexachloro-2,2'-dihydroxy-diphenylmethane (II), or di-(3,5-dichloro-2-hydroxyphenyl) sulphide. A typical composition, which for use can be diluted with ~99 pt. of water comprises I 48.5, II 1.5, Na₂SiO₃ 13, Na₂CO₃ 20, Na dodecylbenzenesulphonate (30% active component) 4, Na tripolyphosphate 10 and water 1%.

F. R. BASFORD.

Substituted umbelliferone esters of thiophosphoric acids and pesticidal compositions containing them. Montecatini Società Generale per l'Industria Mineraria e Chimica (B.P. 811,644, 13.4.56. It., 14.4.55).—Compounds useful in the control of *Musca domestica* (especially those strains resistant to the usual chlorinated insecticides) but of low toxicity to warm-blooded animals comprise dialkyl thiophosphoric acid esters of 7-hydroxy-3,4-trimethylenecoumarin and -3,4-tetramethylenecoumarin (3,4-tri- and -tetramethylene-umbelliferone). The prep. of the diethyl ester of 3,4-trimethylene-coumarin-7-yl thiophosphate m.p. 88—89°, is described.

F. R. BASFORD.

Testing of water for free and combined chlorine. A. T. Palin (B.P. 813,493, 28.11.56).—A sample of the water is added to an indicator consisting of a dialkyl(diethyl)-*p*-phenylenediamine or of a salt thereof, and a sequestering agent consisting of a polyphosphate and/or an aminopolycarboxylic acid, or of a salt thereof (Na₂ EDTA), the amount of the latter being sufficient to minimise the interference from any other substance capable of reacting with the indicator in a manner similar to that of Cl.

E. ENOS JONES.

4.—APPARATUS AND UNCLASSIFIED

Ice machines in yeast production. Anon. (*Industr. Refrig.*, 1960, 138, No. 1, 4).—Five machines producing 25,000 lb. of ice flakes per 24 hr. are arranged to feed directly into the yeast mixing and tempering plant. The ice must be completely frozen, dry and hard and the flakes must be of uniform size. One- and five-lb. blocks of yeast are formed. By using this method the cost of ice has been substantially reduced. If irregular grain size is used there is excess moisture and unequal cooling.

C. V.

[A] **Slit-feeding apparatus for paper chromatography, with examples concerning phosphatides.** [B] **Chromatography of lecithin on non-impregnated paper by means of a slit-feeding apparatus.** H. G. Bungenberg de Jong and J. T. Hoogveen (*Proc. K. Ned. Acad. Wet.*, 1960, 63B, 1—14, 15—19).—[A] In chromatography of phosphatides on silica-impregnated paper, with di-isobutyl ketone/acetic acid/water as mobile phase, the R_F values depend on the ascension height of the front, and the position of the immersion line on the chromatogram is more important than that of the starting spot. A special apparatus was devised, consisting of a glass container, in which is a vertical filling tube, connecting with a horizontal tube at a standard distance from the bottom; this tube has a slit along the top. The paper is marked with an immersion line at 5 mm. from the bottom, and suspended with the lower end in the slit-tube, the line coinciding with the entrance of the slit. The slit-tube is then filled through the vertical tube, with precautions to ensure that the moving front is horizontal. One form of the apparatus takes strips 4.5 × 20.5 cm., and another sheets 17 × 27 cm., allowing a larger no. of spots, and ensuring better separation (because of the larger air vol. in the container). With carefully defined conditions for using and cleaning the apparatus, reproducible chromatograms are obtained.

[B] Lecithin in 0.1% solution was applied to non-impregnated

paper at nine starting points at equal distances apart on a line sloping up from a bottom corner of the sheet. On chromatographing in the large slit-feeding apparatus described in previous abstr., with di-isobutylketone/acetic acid/water (50 : 25 : 5 by vol.), the spots migrated to points on a curve, those from the lowest starting points being nearly in a horizontal line, and those from the higher starting points rising somewhat higher. The R_F values (in the ordinary sense) are thus not constant. The distance of the horizontal line made by the first few spots from the immersion line (a_i) divided by the distance of the front from the immersion line (f_i) was found to be constant, and independent of the ascension height of the front. It is proposed to denote the quotient a_i/f_i by the symbol R_{Fi} . The results suggest that a gradient is present in the mobile phase, and that the horizontal line may indicate a particular composition of the mobile phase at which the upward migration of lecithin reaches its max. velocity. The experimental fact that R_{Fi} is constant may mean that this particular composition is always situated at a constant fraction of the distance from front to immersion line.

M. D. ANDERSON.

Spectrographic analysis of plant material using sodium pyrrolidone-dithiocarbamate for concentration of trace elements. A. Strasheim, D. J. Eve and R. M. Fourie (*J. S. Afr. chem. Inst.*, 1959, 12, 75—80).

—The Na pyrrolidone-dithiocarbamate-metal complexes were obtained from acid solution and dissolved in CHCl₃. Graphite containing Sn, the internal standard, was added to the partially evaporated solution. The dried extract was heated to 450° for ½ hr. and mixed with Li₂CO₃. The sample was burnt to completion in the anode of a D.C. arc with a current of 10 amp. The analysis lines were: Pb 2833.1 Å, V 3185.4 Å, Mo 3170.3 Å, Mn 3044.6 Å, Zn 3345.0 Å, Co 3044.0 and 3453.5 Å, Ni 3050.8 Å, Sn (internal standard) 3032.8 Å. Standards were prepared containing 3—1000 p.p.m. Pb, V, Mo, Co, Ni and 30—10,000 p.p.m. Zn, Mn. The standard deviations from 18 determinations are given and the results compared with those of other workers. Reproducibility is not very good, but the use of Sn rather than Fe as internal standard obviates the accurate determination of Fe in the original sample.

A. ABBOT.

Accurate determination of boron in plant material. R. J. Davidson and W. J. A. Steyn (*J. S. Afr. chem. Inst.*, 1959, 12, 81—86).—The influence of pH, temp. and the drying period on the development of colour in the colorimetric determination of B using curcumin was investigated. Slight modifications were made to the method and a calibration curve was constructed. The precision of the method was tested by carrying out 20 separate determinations on a well-mixed citrus leaf sample containing 109 p.p.m. of B. A % standard deviation of 1.9 was found. P, Ca, Mg, K, S, Fe, Mn, Zn do not interfere. The method has been used for samples containing from 6 to >200 p.p.m. of B.

A. ABBOT.

Effect of chrysanthemum-monocarboxylic acid upon respiratory pathways. R. C. Kestenbaum (*Dissert. Abstr.*, 1959, 20, 1129).—DL-trans-Chrysanthemum-monocarboxylic acid (CMCA) inhibited the growth of many micro-organisms, *Penicillium notatum* being more markedly affected than *Streptococcus faecalis*. Investigation of the effect of CMCA on various enzyme systems showed that it does not exert a primary action on SH groups or S—S bonds, and is not a general protein inactivator. It appears to act on cellular respiration, the probable sites of inactivation being in the cytochrome system, and in the transport of H from flavoprotein to methylene blue.

M. D. ANDERSON.

Micro-determination of selenium in biological materials. R. Handley and C. M. Johnson (*Analyst. Chem.*, 1959, 31, 2105—2106).—Se (0.25—10 µg.) in, e.g., plant tissue, can be determined by means of Cheng's method (*ibid.*, 1956, 28, 1738) in which the yellow SeIV—3,3'-diaminobenzidine complex (I) is utilised. The sample is wet-washed with HNO₃—H₂SO₄—HClO₄ and the mixture is then distilled with HBr—Br in an apparatus (illustrated) ensuring min. vol. of distillate and min. concn. of HBr therein. To the neutralised solution is added HCO₂H to pH 2.5, followed by 0.5% aq. 3,3'-diaminobenzidine and extraction of I with toluene at pH 7. The extinction of I is measured at 420 mµ and 25°. Values in the range 0—5 µg. Se are slightly low.

W. J. BAKER.

Colorimetric determination of selenium in biological materials. C. W. Bonhorst and J. J. Mattice (*Analyt. Chem.*, 1959, 31, 2106—2107).—The sample is digested with HNO₃—H₂SO₄ (containing HgO) and conc. HCl is added to prevent interference by Fe and SO₄²⁻. The cool solution is saturated with SO₂ and, after addition of NH₂OH, is warmed, set aside for 48 hr. and then filtered. The Se⁴⁺ in the filtrate is determined spectrophotometrically by Cheng's method (*ibid.*, 1956, 28, 1738). Accuracy is within 0.2 µg. if pure diaminobenzidine is used (cf. Hoste, *Anal. Chim. Acta*, 1948, 2, 402).

W. J. BAKER.

Journal of Applied Chemistry

The following papers are appearing in the August, 1960, issue

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