CLIV. EXPERIMENTS ON NUTRITION. VIII.
COMPARATIVE VITAMIN B VALUE OF FOODSTUFFS.
CEREALS. I.

By ROBERT HENRY ADERS PLIMMER, JOHN LEWIS ROSEDALE
(*Beit Memorial Research Fellow*), WILLIAM HENRY RAYMOND AND
JOHN LOWNDES.

From the Chemical Department, St Thomas's Hospital Medical School, London.

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It became evident from our previous experiments [Plimmer, Rosedale and Raymond, 1927] that all the constituents of the diet needed balancing by vitamin B to ensure satisfactory rearing of chickens. A similar balance was found in the cases of pigeons and rats. On a basal diet free from vitamin B, large quantities, such as 4 % of dried yeast, or 8 % of marmite, were found necessary to maintain pigeons for long periods.

It is essential to know not only the constituents of the diet which supply vitamin B, but also and more particularly the relative amount of it contained in each foodstuff. Cooper [1912, 1914] and Chick and Hume [1917] have given a most valuable series of comparative data, and there are a few figures by American workers. The figures of Cooper and of Chick and Hume have been expressed in terms of daily quantities on a fixed daily intake of food. From these data figures bearing a relation to the food consumption can be calculated. No quantitative information is available about the common cereals, like whole wheat, rye, maize, though data were given for wheat germ, bran, barley. There is also little quantitative information about vegetables, fruits and other foodstuffs.

These experiments have therefore been undertaken to obtain these facts about the commoner foodstuffs and thus add to our knowledge about food analysis. Some cereals and cereal products have so far been tested and experiments are in progress with other foodstuffs. The figures are purely of relative value. The exact amount of vitamin B in foodstuffs cannot be known till the substance has been isolated and identified, and even then it may not be possible for some time to devise a method for its direct estimation in foodstuffs. Meanwhile feeding experiments are required for information about nutrition.

EXPERIMENTAL METHODS.

The vitamin B value of foodstuffs was tested by Cooper and by Chick and Hume by methods referred to as the curative and the preventive. The former gives information more rapidly and is most useful for testing the value of extracts. The results depend to a considerable extent upon the actual state of the pigeon affected by polyneuritis. Sometimes the birds rapidly reach the condition with retracted head and are easily cured; sometimes this condition is reached more slowly and the cure is more difficult. These peculiarities have also been observed by Kinnersley and Peters [1925]. The preventive method gives more exact information about a foodstuff, as its effect is observed over a considerable period, usually from 60 to 100 days. The pigeons were fed artificially with 30 to 40 g. of white rice per day and a daily quantity of the foodstuff under examination. On account of the laboriousness of the method only a few foods were tested. The results of the two methods have agreed fairly well, but there were some considerable divergences.

Our previous observations [Plimmer and Rosedale, 1923] on two pairs of pigeons on a diet of oatmeal and fishmeal have led us to adopt the preventive method. These birds were maintained for a long period and were allowed to feed *ad lib.* The food consumption amounted to 40 to 45 g. per day. They were not able to rear their young without the addition of marmite to the food. In these experiments instead of artificial feeding we have fed the birds *ad lib.* and we have taken as our standard long maintenance with rearing of young. This standard is much more severe and is possibly too high. At any rate a period of 100 days is sometimes too short. We have often observed maintenance for this time and then the birds have lost their appetite with consequent loss of weight and have died of polyneuritis. Loss of weight always accompanies loss of appetite and at present it is not yet proved that another factor than the antineuritic is concerned in their nutrition.

Foodstuffs can be placed in several groups according to the amount of vitamin B that they contain. Those with none, like white rice and white flour, those very rich, like dried yeast and wheat germ, those with intermediate quantities, which may be much or little.

Our method of testing has been to take a foodstuff, such as whole wheat, to add to it 5 % of fishmeal to supply protein and mineral salts, and see if a pair of birds could be maintained on it with reproduction. If this occurred, then a proportion of white rice (10 %) was added and reproduction once more awaited. Further dilutions of the grain or other food were then made with more white rice, until it was found that the pigeons could no longer rear their young. It was not necessary to add an antiscorbutic to the food, nor has it been found necessary to add cod-liver oil to supply fat-soluble vitamins.

This standard of testing has been found rather too severe, as in some cases there has been maintenance in perfect health without reproduction. It has been observed that if the food is satisfactory the birds eat the rice first; if

not satisfactory, the rice is left. White flour has then been substituted for white rice. This has the advantage that the food mixture can be made into a dough with water and the dough can be made into pills and dried. The birds must then either eat the whole food or leave it. The minimum quantity for maintenance on such a mixture has still to be found in several cases. The results of the experiments are given in a series of tables and additional information is supplied by a short further description.

All the birds have been kept in cages as described previously [1927] on the roof of the building, conditions which are more natural than small cages for one bird or for several. The birds were kept as quiet as possible so as to prevent neglect of their eggs and young ones.

OATMEAL.

The two pairs of adult pigeons, which had already been kept for over a year on oatmeal and fishmeal, were continued on the same diet, with or without the addition of marmite, until the end of 1923. The red pair (Nos. 1 and 2) hatched and reared three pairs of young with marmite; a fourth pair, started with marmite, died on removal of the marmite. The blue pair (Nos. 4 and 5) without marmite had five lots of eggs and failed to rear the young. On including marmite in the food, a pair of young was reared. The details about these pairs of birds are given in Table I.

Table I. *Oatmeal.*

Date	Time in weeks	Diet				Weights, g.		Number of eggs	Result
		Oat- meal	Mar- mite	Cod- liver oil	Fish- meal	Cock No. 1	Hen No. 2		
14. vi. 23		90	1.5	0	5	510	500	6 during June, July, Aug.	4 hatched and reared; 1 cracked; 1 un- fertilized
to								2 in Sept.	Both hatched; 1 squab died on 27th day, no cause of death found. Marmite now omitted and 2nd squab lost weight and died
2. xi. 23	20								
3. xi. 23		90	0	0	5	500	545		
to									
31. xii. 23	9					555	655		
						No. 5	No. 4		
14. vi. 23		90	0	1	5	395	390	10 during June, July, Aug., Sept.	All fertile; 7 died at hatching; 3 hatched and died
to									
17. x. 23	18								
18. x. 23		90	1.5	1	5	445	420	2 in Oct.	Both hatched and reared
to									
31. xii. 23	11					505	430		

The blue pair (Nos. 4 and 5) were continued on the oatmeal diet for the next two years with the results shown briefly in Table II. This pair was used mainly as a control for all experiments with the different cereals.

At first, 1.5 % marmite was added and a pair of young was hatched and reared for a week. The hen then unfortunately escaped and the young were deserted by the cock. With a new hen, after a single egg from which the squab died, two young were reared. Marmite was then raised to 2 %, but the eggs laid were spoilt.

Table II. *Oatmeal.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Oat- meal	Mar- mite	Fish- meal	Cock No. 5	Hen No. 4		
1. i. 24 to 18. iii. 24	11	93.5	1.5	5	480	390	4 in Jan., Feb.	1 broken; 1 unfertile; 2 hatched. Hen escaped; squabs died
						No. 6		New hen
19. iii. 24 to 27. v. 24	10	93.5	1.5	5	460	390	3 in May, June	1 hatched and died; 2 hatched and reared
28. v. 24 to 23. vii. 24	8	93	2	5	425	445	2 in July	Fertile and deserted
24. vii. 24 to 29. iv. 25	40	95	0	5	420	450	2 in Aug. 2 in Feb.	Both hatched; 1 died; 1 reared 1 broken; 1 died at hatching
30. iv. 25 to 24. vi. 25	8				420	375	1 in April	Hen died; egg fertile and deserted. Cock only
					400			
25. vi. 25 to 17. xii. 25	25	95	0	5	400	380	12 during June, Aug., Sept., Oct., Nov.	New hen 3 unfertile; 3 hatched and reared, but poor growth and squabs unwell; 2 deserted; 2 dead in shell; 2 hatched and died
					540	510		

It was desired to test oatmeal alone over a long period. In 48 weeks only two lots of eggs were laid, from which only one squab was reared. After 6 months two more eggs were laid but young were not reared. A single egg was then laid and deserted. The hen was then unwell and died. The cock was also losing weight at this period. With a new hen (No. 4) one young was reared from the first two eggs, both were reared from the next eggs, and subsequently no more were reared.

Adult pigeons can thus be maintained for long periods on a diet of oatmeal and young can occasionally be reared. Young can generally be reared with an addition of marmite (2 %). The long period on oatmeal caused a slight loss of weight of the adult birds, the hen being more affected than the cock, possibly from loss of vitamin B from her body into the eggs. One hen died on the long period of oatmeal feeding.

Oatmeal would thus appear to contain just enough vitamin B for maintenance of pigeons, but not enough for rearing.

SUSSEX GROUND OATS.

The red pair of pigeons (Nos. 1 and 2) was changed over to Sussex ground oats on 1. i. 1924. The hen laid two lots of eggs in the first period of 20 weeks, but no young were reared. In the next two periods, amounting together to 21 weeks, marmite was added. Young ones were again not reared. The cause of non-rearing seemed due rather to the fibrous nature of the food than to lack of vitamin B. The young thrived for a time and died with fibre in the gizzard. At the end of the breeding season the adult birds were kept a final 11 weeks on Sussex ground oats and both increased in weight.

Sussex ground oats are thus similar to oatmeal. Adult pigeons can be maintained, but young cannot be reared.

The details are shown briefly in Table III.

Table III. *Sussex ground oats.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Sussex ground oats	Mar- mite	Fish- meal	Cock No. 1	Hen No. 2		
1. i. 24 to 20. v. 24	20	95	0	5	495	600	4 in Mar., Apr.	3 died after hatching; 1 fertile, not hatched
21. v. 24 to 18. vii. 24	9	91	4	5	435	490	4 in May, June	1 unfertile; 3 hatched and died
19. vii. 24 to 13. x. 24	12	89	6	5	465	510	4 in July, Aug.	2 addled; 2 hatched and died in 1st week
14. x. 24 to 31. xii. 24	11	95	0	5	440	495		
					430	510		

WHOLE OATS.

The pair was changed over to whole oats for a period of one year (1925). The change had a favourable effect upon the rearing of young. Both young from the first pair of eggs were reared, one from the second lot, and one from the fourth lot. The first young ones were practically normal, but the later ones showed poor growth and were not first-class birds on removal from their parents. The adult pair maintained their healthy appearance throughout, but increased in weight too much at the end. The difference in result between Sussex ground oats and whole oats is probably due to fibre. The husk of the whole oat was usually discarded by the birds, whilst such separation was not possible from the ground oats.

Whole oats appear rather better than oatmeal. In the preparation of oatmeal, a portion of the germ with vitamin B seems to be removed.

The details are shown in Table IV.

Table IV. *Whole oats.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Whole oats	Mar- mite	Fish- meal	Cock No. 1	Hen No. 2		
1. i. 25		95	0	5	430	510	8 in Mar., May, June, July	4 hatched and reared, poor growth never quite normal; 2 added; 2 hatched and died in 2nd week
to								
30. xii. 25	52				535	655		

BARLEY.

This pair of birds (Nos. 21 and 22) was given barleymeal for the first year (Table V). The fertility of the eggs was not good and changes in the diet had to be made after the rearing of a single squab. No squabs were produced with the diet of 70 % barleymeal. On this quantity the pair was kept for 20 weeks: no loss in weight of the birds occurred.

Table V. *Barley.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Barley- meal	Rice	Fish- meal	Cock No. 21	Hen No. 22		
1. i. 24		90	0	5	420	460	6 in Mar., Apr., May	2 unfertile; 2 hatched and reared; 1 fertile, des- erted; 1 hatched and died
to								
1. vii. 24	25							
2. vii. 24		80	10	5	395	415	2 in June, July	1 unfertile; 1 hatched and reared
to								
12. viii. 24	7							
		Barley grains						
13. viii. 24		70	20	5	420	495	4 in Aug., Sept.	2 added; 2 unfertile
to								
31. xii. 24	20							
1. i. 25		70	20	5	420	500	2 in Jan.	Deserted, unfertile. Cock lost
to								
10. ii. 25	7				No. 121 410	480		New cock
to								
13. iv. 25	9						2 in Feb.	2 hatched and reared
14. iv. 25		60	30	5	410	470	8 in Apr., May, July, Aug.	1 unfertile; 1 added; 6 hatched and reared
to								
4. v. 25	3	63.5	31.5	5				
to								
8. vi. 25	5	65	30	5				
to								
5. x. 25	18							
6. x. 25		55	40	5	420	465	6 in Jan., Feb., Mar.	4 broken and deserted; 1 died at hatching; 1 died in 2nd week with symptoms of polyneu- ritis. Hen died, blood clot near heart
to								
26. iii. 26	24				380	480		
					No. 25/34			New hen
2. iv. 26					390	400	6 in May, June, Aug.	3 hatched and reared; 1 dead in shell; 2 fertile, and deserted
to								
20. i. 27	42				480	500		

During the second year barley grains were substituted for barleymeal. The birds had experienced some difficulty in picking up the fine meal and consequently complete consumption of the food was not really attained. With grains of barley, it was observed that all the food was picked up and eaten.

Unfortunately the cock escaped soon after this change. With a new cock, the fertility of the eggs was improved and a pair of young was reared on 70 % barley.

On reducing the barley to 60 %, then raising to 63·5 % and later to 65 %, squabs were hatched and reared.

Reduction of the barley to 55 % did not allow of rearing. One squab which was hatched died with symptoms of polyneuritis. Subsequently the hen died. With a new hen, altogether three squabs were reared—the next eggs were fertile and deserted and then no more eggs were laid. This condition of maintenance without reproduction has been observed in other cases.

55 % of barley thus seems the limiting quantity for rearing, but 65 % is better. It will be necessary to ascertain the limiting quantity for maintenance in future experiments.

RYE.

Whole rye seeds were used as food for these birds (Nos. 9 and 10). As shown in Table VI, the birds were first given rye and fishmeal. After the rearing of

Table VI. *Rye.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Rye	Rice	Fish- meal	Cock No. 10	Hen No. 9		
5. iii. 24 to 27. v. 24	12	95	0	5	375	415	2 in Apr.	Both hatched and reared
28. v. 24 to 8. vii. 24	6	80	10	5	390	420	2 in May	Both hatched and reared
9. vii. 24 to 22. iv. 25	41	70	20	5	430	435	12 in July, Aug., Sept., Oct., Jan., Mar.	10 fertile, deserted; 2 hatched and reared
23. iv. 25 to 10. vi. 25	7	60	30	5	390	415	2 in May	Both hatched and reared
11. vi. 25 to 9. ix. 25	13	55	40	5	450	425	4 in June, July	1 addled; 1 died at hatching; 2 hatched and reared
10. ix. 25 to 14. iv. 26	31	45	50	5	400	425	2 in Feb.	Both hatched and reared
15. iv. 26 to 7. vii. 26	12	40	55	5	400	405	2 in Apr.	1 fertile, deserted; 1 hatched and reared. Birds do not eat rice
8. vii. 26 to 22. ix. 26	11	45	50	5	345	370		Birds still leave rice
23. ix. 26 to 24. xi. 26	9	50	45	5	360	395		Rice eaten; birds better

a pair of squabs, rice was added in gradually increasing quantities according as young were reared. The birds were kept for the long period of 41 weeks with 70 parts of rye, mainly on account of the eggs being deserted. This was largely due to the very wet season; the nest of the birds was continually swamped with water. On reducing the rye to 60 parts, young were hatched from the first pair of eggs. Young were hatched and reared with as little as 45 % of rye. With less rye in the diet, the adult birds after a time did not eat the rice and began to look unwell. The quantity was obviously too little. No great improvement in the birds was seen on raising the rye to 45 %, but on giving 50 % rye the birds improved. It can be concluded that adult birds require 45 % rye for maintenance and that 50 to 55 % is required for rearing of young. It will be of interest in future experiments to prove definitely whether a minimum of 40 or 45 % is needed for maintenance, which will correspond with the figure found for wheat.

WHEAT.

Whole wheatmeal flour, such as is used for human consumption, was given as the food for these birds (Nos. 17 and 18) (see Table VII). At first, the flour was wetted with water to bind the fine particles together, but later, on mixing with rice, the two components (the rice finely ground) and the fishmeal were made into a dough, and the dough was made into small pills which were dried on glass plates at room temperature or at 37°. Two young were reared up to a proportion of 70 % wheat. Many eggs were laid on the diet with 60 % wheat during the long period of 31 weeks, but no young were hatched. The proportion of whole wheat was then raised first to 70 % and then to 75 %. Again young were not hatched out. The hen then died. No particular symptoms could be found at *post mortem* examination and it seemed that she died of exhaustion from laying so many eggs, 30 in all.

With a new hen, keeping the whole wheat at 75 %, during the long period of 51 weeks, only two young were hatched and reared. Two other pairs of young were hatched, but they died in the second week with symptoms of polyneuritis. The other eggs were fertile, but no young were hatched from them.

It thus appeared that 75 % whole wheat was the minimum for the hatching and rearing of young, but that 80 % was distinctly better.

The quantity of whole wheat in the diet was now reduced to 60 %. The eggs which were laid were fertile, but always deserted.

In the meantime another pair of birds had been kept on 50 % whole wheatmeal (see below) and it was decided to find out what was the minimum quantity of whole wheatmeal required for the maintenance of adult birds. The quantity of whole wheatmeal was therefore reduced to 30 %. On this quantity both birds died, which showed definitely that more than 30 % was needed.

Another pair of birds (Nos. 25/2 and 25/3) was kept on 50 % whole wheat-

meal, which was mixed with white flour instead of rice, for 45 weeks. One young was hatched and reared from the first pair of eggs. The other eggs were fertile and were deserted. To test whether an increase of whole wheat-meal flour to 75 % would improve the hatching and rearing, the birds were given this diet for another 34 weeks. Rather more eggs were laid, but they were always deserted. During the time on 50 % whole wheat the birds kept a constant weight. This quantity thus seemed a satisfactory amount for long maintenance.

Table VII. *Wheat.*

Date	Time in weeks	Diet		Fish- meal	Weights, g.		Number of eggs	Result
		Wheat	Rice		Cock No. 17	Hen No. 18		
26. iii. 24 to 28. v. 24	10	90	0	5	440	365	4 in Apr., May	2 hatched and reared; 2 deserted
29. v. 24 to 23. vii. 24	8	80	10	5	430	370	2 in June	Both hatched and reared
24. vii. 24 to 3. ix. 24	6	70	20	5	425	365	4 in July, Aug.	2 hatched and reared; 2 deserted
4. ix. 24 to 7. iv. 25	31	60	30	5	440	365	14 in Sept., Oct., Nov., Jan., Feb., Mar., Apr.	4 unfertile; 10 added and deserted
8. iv. 25 to 6. v. 25	4	70	20	5	450	385	2 in May	1 unfertile; 1 fertile, de- serted
7. v. 25 to 17. vi. 25	6	75	20	5	445	380	4 in May, June	1 very small egg; 2 un- fertile; 1 added. Hen died; no particular symptoms
1. vii. 25 to 26. vi. 26	51	75	20	5	No. 118 400	335	12 in July, Oct., Feb., Mar., May	New hen 1 added; 2 hatched and reared; 2 hatched and died in 2nd week with enlarged hearts; 4 de- serted; 2 dead in shell; 1 hatched, died in 2nd week with polyneuritis
27. vi. 26 to 29. ix. 26	14	60	35	5	430	365	7 in June, July, Aug., Sept.	5 deserted, fertile; 2 dead at hatch
30. ix. 26 to 7. iii. 27	23	30	65	5	450	415	2 in Oct.	Deserted, fertile. Hen died on 31. xii. 26 after 13 weeks. Cock died of polyneuritis
18. xi. 25 to 29. ix. 26	45	50	45	5	350 No. 25/2 No. 25/3 440	445	8 in Feb., Apr., June, July	1 dead in shell; 1 hatched and reared; 1 hatched and died; 5 fertile and deserted
30. ix. 26 to 25. v. 27	34	75	20	5	415	465	9 in Oct., Feb., Mar., Apr., May	8 fertile and deserted 1 unfertile
10. iv. 27 to 17. viii. 27	23	40	55	5	435 No. 93 No. 26/27 455	460 380	6 in Apr., May, June	1 hatched and reared, but very poor growth; 2 un- fertile; 3 fertile and deserted

It remained to ascertain if 40 % whole wheatmeal would be enough for maintenance. Another pair of birds (Nos. 93 and 26/27) was started on this quantity. They managed to rear one young one from the first lot of eggs, but this young one showed very poor growth and was very small. Two other lots of eggs were laid which were deserted and then no more. They have been kept for over 20 weeks without signs of illness, but they have shown a slight loss in weight.

It may be concluded that 40 % of whole wheat is the minimum for maintenance of adult pigeons, 50 % is a more favourable quantity and 75 to 80 % whole wheat is required for reproduction and rearing of young.

WHEAT GERM.

The high vitamin B value of wheat germ was determined by Chick and Hume [1917]. They found that pigeons on a daily diet of 30 to 40 g. of white rice were protected over a period of 63 days by a dose of 3 g. of wheat germ given every second day. This amount corresponds to about 4.5 % of the diet. According to our experiments the smallest quantity which must be present in the diet is 6 %. Table VIII shows our results. The first period was for

Table VIII. *Wheat germ.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		White rice	Wheat germ	Fish- meal	Cock No. 24/13	Hen No. 24/10		
23. vii. 24 to 11. xi. 24	16	90	10	5	360	330		
12. xi. 24 to 17. ii. 25	14	90	6	5	445	395		Birds found to be two hens
18. ii. 25 to 30. vi. 25	19	White flour 89	6	5	No. 31 460	No. 24/10 440	8 in Feb., Mar., Apr., May, June	1 unfertile; 2 addled; 2 dead at hatching; 3 fer- tile and deserted
1. vii. 25 to 20. xi. 25	20	87	8	5	425	385	6 in July, Aug.	All deserted. 24. viii. 25, hen ill, dosed with 4 doses of 2 g. wheat germ; better. Hen died with polyneuritis; en- larged heart New hen
25. xi. 25 to 1. vi. 26	27	85	10	5	460	No. 25/31 460	4 in Mar., Apr.	Fertile and deserted
2. vi. 26 to 27. vii. 26	8	93	2	5	430	395	1 in June	Fertile, deserted. Hen died 7. vii. 26, in 5 weeks. Cock died
8. vii. 26 to 22. ix. 26	11	91	4	5	No. 26/2 345	No. 26/1 395	2 in July	Deserted. Both birds died of poly- neuritis

16 weeks with a proportion of 10 % of wheat germ and was followed by a period of 14 weeks with 6 %. The supposed cock of this pair of birds was then discovered to be a hen. On exchanging one of the hens for a cock the pair was kept for 19 weeks on 6 % of wheat germ. At this time white flour was used instead of white rice, and the diet given in small pills.

The hen laid eggs during this period, but they were always deserted. The quantity of wheat germ was then raised to 8 % to see if any improvement in hatching would take place. The hen showed signs of polyneuritis after 8 weeks, but was cured with four doses of 2 g. of wheat germ. At 20 weeks the hen again suffered from polyneuritis and died.

Another hen was placed with the cock and the amount of wheat germ was raised to 10 %. The hatching was no better and after 27 weeks the attempt to produce rearing was discontinued.

It was decided to find out the smallest quantity of wheat germ which would suffice to maintain a pair of pigeons. The quantity was therefore reduced to 2 %. Both birds died in 8 weeks.

Table IX. *Heated wheat germ.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		White rice	Heated wheat germ	Fish- meal	Cock No. 24/12	Hen No. 24/14		
23. vii. 24		90	10	5	315	370		Cock lost
to					No. 9131			New bird
11. xi. 24	16				430	440		
12. xi. 24		90	6	5	365	430		New bird found to be hen
to								
11. ii. 25	13				450	450		
to					No. 25			Another cock
4. iii. 25	3				380	410		Hen died from unknown cause
					390	395		
		White flour			No. 24/23			New hen
11. iii. 25		89	6	5	375	370	6 in Apr., May, June	4 fertile and deserted; 1 added; 1 died at hatching
to								
30. vi. 25	16							
1. vii. 25		87	8	5	405	395	10 in July, Aug., Sept., Oct.	2 deserted; 1 unfertile; 5 hatched and died; 2 added
to					430	430		
18. xi. 25	20							
19. xi. 25		85	10	5			14 in Nov., Dec., Jan., Feb., Mar., Apr.	5 unfertile; 3 fertile and deserted; 5 broken; 1 added
to					390	400		
12. v. 26	25							
13. v. 26		83	12	5			2 in May, June	1 egg only at each time of laying; both broken
to								Cock escaped
28. vii. 26	10				No. 26/6	No. 26/3		
27. vii. 26		91	4	5	405	385		Cock died in 6th week; hen died in 8th week
to								
22. ix. 26	8							

Another pair was started on 4 % wheat germ. These died in 11 weeks. 2 % and 4 % wheat germ were therefore insufficient to balance the white flour. On 6 % two birds were kept for 14 weeks and another pair for 19 weeks. One of the two had been for both these periods on this quantity. 6 % must therefore be taken as the minimum quantity for long maintenance.

HEATED WHEAT GERM.

At the same time as the experiments with wheat germ were being made, similar ones with heated wheat germ were carried out. The wheat germ is heated to about 120° to prevent its decomposition. Table IX shows the results. A pair of birds was kept for 16 weeks with 6 % heated wheat germ. There was a slight improvement in hatching with 8 and 10 % wheat germ.

A pair of birds on 4 % of heated wheat germ died in 8 weeks. For maintenance therefore 6 % of wheat germ is required. The germ does not lose vitamin B by this treatment¹.

BRAN AND MIDLINGS.

The high content of vitamin B in bran was found by Chick and Hume [1917]. It appeared to be about one-fifth of that of wheat germ.

Table X. *Bran.*

Date	Time in weeks	Diet				Weights, g.		Number of eggs	Result
		Bran	White flour	Fish- meal	Mar- mite	Cock No. 24/4	Hen No. 24/6		
9. vii. 24 to 2. ix. 24	8	45	45	5	0	365	365		
3. ix. 24 to 14. iv. 25	32	30	60	5	0	395	455	6 in Jan., Feb., Mar.	4 died at hatching; 1 addled; 1 fertile, deserted
15. iv. 25 to 22. ix. 25	23	47.5	47.5	5	0	430	425	10 in Apr., May, June, July, Aug., Sept.	3 died at hatching; 2 unfertile; 2 addled; 3 hatched and died
23. ix. 25 to 2. ii. 26	19	33	60	5	2	420	450	4 in Oct., Jan.	2 hatched, 1 died 1 reared; 2 fertile and deserted
3. ii. 26 to 18. v. 26	15	33.3	57.7	5	4	430	455	4 in Feb., Apr.	2 unfertile, deserted; 2 addled, deserted
19. v. 26 to 22. vi. 26	5	33.3	55.7	5	6	385	410	2 in May	Deserted
					Dried yeast				
23. vi. 26 to 29. ix. 26	14	33.3	55.7	5	6	380 415	400 425	5 in July, Sept.	1 egg only and de- serted; 4 fertile and deserted

¹ The wheat germ and heated wheat germ were kindly supplied by Mr W. T. Ryle. Messrs Hovis, Ltd., have also kindly given us wheat germ for these experiments.

Table XI. *Middlings.*

Date	Time in weeks	Diet				Weights, g.		Number of eggs	Result
		Midd- lings	White rice	Fish- meal	Mar- mite	Cock No. 24/3	Hen No. 24/5		
9. vii. 24 to 2. ix. 24	8	70	20	5	0	370	335		
3. ix. 24 to 9. xii. 24	14	60	30	5	0	365	355		
10. xii. 24 to 31. iii. 25	16	30	60	5	0	415	385	4 in Feb., Mar.	1 unfertile; 1 addled; 2 dead at hatching
		White flour							
1. iv. 25 to 16. vi. 25	11	30	60	5	0.6	435	380	5 in Apr., May, June	1 deserted; 2 addled; 1 unfertile; 1 died at hatching
17. vi. 25 to 13. i. 26	30	33.3	60	5	1.7	405	375	10 in June, July, Aug., Sept., Nov.	4 deserted; 2 hatched and reared; 4 hatch- ed and died
14. i. 26 to 5. v. 26	16	33.3	58.7	5	3.0	495	440	8 in Jan., Feb., Mar., Apr.	2 dead at hatch; 6 broken, deserted
6. v. 26 to 23. vi. 26	7	33.3	55.7	5	6.0	435	375	4 in May, June	Broken and deserted
					Dried yeast 6.0				
24. vi. 26 to 29. ix. 26	14					395	335	2 in July	Broken. Hen ill 30. viii. 26
						430	360		

In order to obtain further comparative figures for these foodstuffs pairs of pigeons were started on 45 % bran and on 70 % middlings. As shown in Tables X and XI, the pair was kept on 30 % bran for 32 weeks. As they were not able to rear young the amount of bran was raised to 47.5 %. Some eggs were hatched, but the young died. On account of the large amount of fibre in bran, extra vitamin B was supplied by incorporating a proportion of marmite; one young squab was then reared. With a further increase of marmite, no more young were reared and there was no improvement on changing from marmite to dried yeast. The attempt to rear was then abandoned.

Similar results were obtained with middlings. There was better hatching and rearing on adding marmite to the diet. With still more marmite, there was no better rearing. It would appear that some other factor than vitamin B was now concerned in reproduction. Both with bran and middlings the weights of the parent birds remained practically constant.

As far as maintenance of adult pigeons is concerned, the pair was kept on 30 % bran for 32 weeks, and the pair on 30 % middlings for 16 weeks. In future experiments it is proposed to ascertain the smallest quantities of bran and middlings upon which adult pigeons can be maintained for 20 to 26 weeks.

MAIZE.

Kibbled maize (see Table XII) was used as food for these pigeons (Nos. 13 and 14). At first, partly owing to the wet weather, the eggs were deserted. An addition of marmite was made to the food and ultimately a squab was reared.

Table XII. *Maize.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Kib- bled maize	Mar- mite	Fish- meal	Cock No. 14	Hen No. 13		
19. iv. 24 to 27. vi. 24	10	90	0	5	445	385	6 in Apr., May, June	4 fertile and deserted; 2 added
28. vi. 24 to 8. viii. 24	9	90	2	5	455	400	2 in July	Added
9. viii. 24 to 24. x. 24	11	90	3	5	465	420	2 in Sept.	1 added; 1 hatched and reared
25. x. 24 to 29. v. 25	31	90	0	5	500	440	14 in Oct., Nov., Dec., Jan., Mar., Apr.	4 spoilt by wet weather; 4 fertile and deserted; 1 added; 5 hatched and reared
30. v. 25 to 4. ix. 25	14	85	White rice 5	5	435	395	8 in June, July, Aug.	4 hatched and reared; 2 deserted; 2 hatched and died
5. ix. 25 to 30. ix. 26	56	75	20	5	470	415	23 in Oct., Nov., Jan., Feb., Mar., Apr., May, June, July, Aug.	6 hatched and reared; 6 unfertile; 4 added; 7 fertile and deserted
1. x. 26 to 9. iii. 27	23	50	45	5	450	410	16 in Oct., Nov., Dec., Jan., Feb.	6 fertile and deserted; 10 broken
10. iii. 27 to 1. vi. 27	9	95	0	5	495	425	7 in Mar., Apr., May	All broken
29. vii. 26 to 16. iii. 27	33	60	35	5	410	355	6 in Jan., Feb., Mar.	3 fertile and deserted; 1 hatched and died; 2 dead at hatch
17. iii. 27 to 20. vii. 27	18	60	White flour 35	5	435	390	7 in Apr., May, June, July	5 fertile and deserted; 1 fertile and dead at hatch; 1 hatched and died

On returning to the simple maize and fishmeal diet, several young were hatched and reared. The maize thus appeared to contain enough vitamin B for rearing. After adding white rice to the diet, young were hatched and

reared. The pair was kept on 75 % kibbled maize for the long period of 56 weeks during which time 6 squabs were reared. This proportion of maize thus seemed the limit for rearing. No young were reared on 50 % maize, though the parents were kept on it for 23 weeks. On returning to kibbled maize only, rearing was not improved. After the long period of over two years the experiment with these birds was not continued. Lack of the power of rearing was probably due to exhaustion of some other factor which is required.

Another pair of pigeons was in the meantime tried on 60 % kibbled maize. Though many eggs were laid, and some hatched out, there was no rearing of the young.

75 % maize must be regarded as the minimum for rearing. So far 50 % has been found enough for maintenance, but 60 % was distinctly better. Future experiments are designed to find out if pigeons can be maintained on less.

MAIZE GERM-MEAL AND MAIZE GLUTEN.

These two cereal products are often used in feeding poultry and other animals.

The high vitamin B value of maize germ, picked by hand from the grain, has been shown by Chick and Hume [1917]. It was consequently expected that maize germmeal would be rich in vitamin B.

A pair of pigeons was started on 90 parts rice, 10 parts maize germ-meal and 5 parts fishmeal. They lost weight at once and looked ill. On raising the maize germ-meal to 20 parts, there was no improvement and the birds died in altogether 11 weeks.

On trying a pair of pigeons on 90 parts maize germ-meal and 5 parts of fishmeal, they became ill and died in 6 weeks.

Another pair was tried with 90 parts maize germ-meal, 90 parts white flour and 5 parts fishmeal. They also became ill, but recovered on adding 5 % of marmite to the mixture. On reducing the marmite to 3 parts, the quantity to balance the white flour, they became ill and died.

Commercial maize germ-meal thus appears to contain no, or very little, vitamin B.

Maize gluten was not expected to contain vitamin B. The following experiments showed the presence of a small amount. A pair of birds was given 45 parts maize gluten, 45 parts white flour, 5 parts fishmeal and 5 parts marmite. They showed growth for 6 weeks, but on reducing the marmite to 3 parts they lost weight and died.

A pair of birds on 90 parts maize gluten and 5 parts fishmeal died in 4 and 14 weeks.

Maize gluten may thus contain small amounts of vitamin B.

BUCKWHEAT.

Buckwheat, often used in America for human consumption and in France for feeding poultry, has been tested on the pair of pigeons Nos. 23 and 24

(Table XIII). After 18 weeks on buckwheat alone, a pair of squabs was reared. One young one was reared on 80 parts of buckwheat. The hen subsequently laid no more eggs. The pair was maintained without loss of weight for 31 weeks. The cock then died. With a new cock there was no improvement, even after returning to buckwheat alone as the diet. Finally a small proportion of cod-liver oil was added; eggs were still not laid by the hen. This grain seems to differ from the other grains by not permitting the laying of the eggs by the hen.

Further experiments are needed to ascertain the peculiarities of buckwheat.

Table XIII. *Buckwheat.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Buck- wheat	White rice	Fish- meal	Cock No. 23	Hen No. 24		
20. ii. 24 to 25. vi. 24	18	90	0	5	420	285	7 in Mar., Apr., May	1 little egg; 2 shell-less; 1 hatched and died; 3 hatched and reared
26. vi. 24 to 5. viii. 24	6	80	10	5	415	300	2 in June	1 unfertile; 1 hatched and reared
6. viii. 24 to 11. iii. 25	31	70	20	5	450	330	No eggs	Cock died; tumour
12. iii. 25 to 16. ix. 25	35	95	0	5	365	340	No eggs	New cock
17. ix. 25 to 6. xii. 25	12	94.5	0	5	400	335	No eggs	With 0.5 % cod-liver oil
					435	345		

MILLET AND DARI.

These two grains much used as human food by the natives of South Africa with good results showed a high vitamin B value on testing on pigeons. Tables XIV and XV show the behaviour of the pigeons. On gradually reducing the proportion of millet in the diet, very good hatching and rearing took place with 60 parts of millet. With 50 parts of millet the rearing was less good, but was slightly improved with 55 parts. The minimum for rearing can be said to be 55 %. Smaller quantities to determine the figure for maintenance have not yet been tried. The birds have been quite well on 50 % for 18 weeks.

The pair of birds on dari were quite prolific, but the fertility of their eggs was not good. A pair of squabs was reared on 50 %. The quantity for rearing is more certain at 60 %.

The maintenance quantity has not been determined, but experiments for this purpose will be undertaken shortly. The pair of birds lived quite well on 50 % dari.

Table XIV. *Millet.*

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Millet	White rice	Fish- meal	Cock No. 15	Hen No. 16		
1. v. 24 to 25. vi. 24	8	90	0	5	350	380	2 in May	Both hatched and reared
26. vi. 24 to 26. viii. 24	9	80	10	5	375	365	4 in June, July	2 addled; 2 hatched and reared
27. viii. 24 to 22. x. 24	8	70	20	5	415	395	2 in Aug., Sept.	1 dead in shell; 1 hatched and reared
23. x. 24 to 31. xii. 24	10	60	30	5	430	400	2 in Nov.	Both hatched and reared
1. i. 25 to 5. v. 25	18	50	40	5	480	425	6 in Jan., Mar., Apr.	1 dead in shell; 1 unfertile; 2 hatched and reared; 2 hatched and died
6. v. 25 to 9. vi. 25	5	53	42	5		420	2 in May	1 dead in shell; 1 hatched and died
10. vi. 25 to 12. x. 25	18	55	40	5	395	380	6 in June, July, Sept.	4 dead in shell; 1 hatched and reared; 1 unfertile
13. x. 25 to 23. xi. 25	6	60	35	5	435	430	2 in Oct.	Both hatched and reared
24. xi. 25 to 25. i. 26	9	55	40	5	440	435	2 in Dec.	Both hatched and reared
					495	430		

BAKER'S YEAST.

Our experience with dried yeast and marmite led to testing the value of baker's yeast. Analysis of a sample showed 2.23 % of N (= 13.9 % of protein), 1.9 % of ash and 71.3 % of water, which would indicate a value of about one-third of dried yeast.

An experiment was started with 10 % of baker's yeast. The birds at first increased slightly in weight, then remained constant for about 20 weeks. During this time the hen laid three lots of eggs. They were fertile, but were deserted. Subsequently, both birds lost their appetite and their weight and died at the end of the 26th week with the typical retracted head symptoms of polyneuritis. Table XVI gives the further information.

Another experiment was made with 4 % of baker's yeast, the amount corresponding with the necessary amount of dried yeast. The birds did not like the food and scattered it. They lost weight at once and both showed polyneuritis in the fifth week. The cock was cured by a dose of 2 g. of wheat germ, but the hen was allowed to die.

Table XV. *Dari*.

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		Dari	White rice	Fish- meal	Cock No. 7	Hen No. 8		
20. ii. 24 to 23. vi. 24	18	90	0	5	400	420	7 in Mar., Apr., May, June	2 unfertile; 2 fertile and deserted; 2 deserted; 1 hatched and reared
24. vi. 24 to 8. viii. 24	8	80	10	5	475	380	2 in July	Both hatched and reared
19. viii. 24 to 6. x. 24	7	70	20	5	490	395	2 in Aug.	1 died at hatching; 1 hatched and reared
7. x. 24 to 17. xi. 24	6	60	30	5	490	435	2 in Oct.	1 died at hatching; 1 hatched and reared
18. xi. 24 to 6. iv. 25	20	50	40	5	570	440	6 in Dec., Jan., Feb.	3 unfertile; 1 fertile and deserted; 2 hatched and reared
7. iv. 25 to 8. vi. 25	9	45	45	5	520	415	4 in Mar., May	3 hatched and reared; 1 unfertile
9. vi. 25 to 30. vi. 25	3	50	45	5	450	400	2 in June	Both died at hatching
1. vii. 25 to 13. x. 25	15	55	40	5	440	380	6 in July, Aug., Sept.	1 hatched and reared; 1 unfertile; 2 addled; 2 dead at hatching
14. x. 25 to 24. xi. 25	6	60	35	5	500	440	2 in Oct.	Both hatched and reared
25. xi. 25 to 5. i. 26	6	55	40	5	530	410	2 in Dec.	Both unfertile
					590	450		

Table XVI. *Baker's yeast*.

Date	Time in weeks	Diet			Weights, g.		Number of eggs	Result
		White flour	Baker's yeast	Fish- meal	Cock No. 53	Hen No. 48		
16. ii. 27 to 17. viii. 27	26	85	10	5	415 440 400 410 340	320 365 320 335 220	6 in Apr., May, June	All fertile and deserted. Both birds died of polyneuritis

DISCUSSION AND SUMMARY.

At the time that these experiments were started, it was generally understood that the term vitamin B applied to the antineuritic substance preventing beri-beri. Yeast and other foods have sometimes been described as possibly containing two unknown food factors, the one antineuritic and the other maintaining body weight and necessary for growth. The work of Goldberger and his colleagues, and, more recently, that of Chick and Roscoe [1927] again tend to differentiate two factors in yeast and wheat embryo. The one factor

is definitely antineuritic and can be destroyed by heating at 120° under pressure; the other is thermostable, preventive of pellagra and maintains body weight and is called by Goldberger the PP factor. Chick and Roscoe wish to call this PP factor vitamin B. The nomenclature of the various unknowns in nutrition is already sufficiently complicated, *e.g.* two D vitamins have been described; the one water-soluble in yeast (Funk) and the other fat-soluble, now more definitely called vitamin D. Funk's vitamin D may be the same as the PP factor. It would have been simpler to leave the term vitamin B for the antineuritic substance with which it has so long been associated and give the term PP factor to the unknown substance in yeast and other foods which prevents pellagra and maintains body weight.

The term antineuritic limits the symptoms to those of paralysis, but a deficiency of this factor is responsible for a whole sequence of other symptoms relating to the alimentary tract, the endocrine organs and the heart, as shown by McCarrison, and observed by us on feeding with too little yeast [1927]. The animal grows, showing the presence of the second factor, but dies with alimentary, heart, or paralytic symptoms according to the degree of shortage of the vitamin.

The existence of two forms of beri-beri, the wet and the dry, has long been known, and it has often been suggested that they were due to two factors. Vedder was never able to produce the dropsical condition in birds, even with extra salts in the diet. Our observations again point to the different forms as being due to the degree of shortage of vitamin B. We have often seen wet tissues in our birds (more often in chickens) when the degree of shortage is slight and exists over a long period. A great shortage causes an early death and the tissues are dry. Before death the bird has generally stoppage of the cloaca. If excretion should occur, the birds get better for a time. If a bird is cured with a dose of yeast or wheat germ, there is first of all excretion and then recovery.

With reference to the unknown factor concerned in preventing pellagra, it is not definitely proved that it is not a constituent of proteins, as Goldberger originally believed. The experimental work of Chick and Roscoe was done with caseinogen, a protein which cannot be looked upon as a complete one. Caseinogen is deficient in cystine and may also lack other protein units. These deficiencies are probably compensated for by lactalbumin in milk, which has been shown to be so efficacious in preventing and curing pellagra and also promotes growth. Yeast protein may contain the same unit and similarly wheat embryo, especially the less pure samples which are better for pellagra prevention and for maintaining body weight. It would be of great interest to know if the same results are obtained with lactalbumin, or other complete protein. The protein in our experiments has been fishmeal.

In any case we hope that Chick and Roscoe will consider our plea to continue the association of the term vitamin B with the antineuritic factor and adopt the term PP factor for the unknown factor in yeast concerned in

pellagra prevention and in maintaining body weight. McCollum and Davis associated the antineuritic substance with vitamin B more particularly from the fact that impure lactose contained vitamin B and would cure polyneuritis in pigeons, rather than from the similar distribution of the substances.

Our results are considered entirely in respect of the antineuritic substance. We have observed previously [1927] that young birds and rats need more of it than adult animals. These data with cereals support those observations. Adult pigeons can be maintained on a smaller quantity of cereal than that upon which young can be reared. The same argument can be applied to the PP factor, that young animals want more than adults, but against this contention is the fact that the young bird grows quite rapidly and then dies showing symptoms of polyneuritis, so that it appears to have enough PP factor. Our observations have also always been to the effect that the bird, whether young or adult, first loses its appetite with corresponding loss of weight and then dies of polyneuritis. The quantity of antineuritic factor is the determining consideration.

Our data may be summarised very simply as follows:

			Percentage quantity of foodstuff required for	
			Rearing	Maintenance
Oatmeal	...	...	More than 95	95
Sussex ground oats	...	...	More than 95	95
Whole oats	...	...	95	95
Barley (whole)	...	...	65	55
Rye (whole)	...	...	50-55	45
Whole wheat flour	...	...	75-80	40-50
Wheat germ	...	...	More than 10	6
Bran	...	...	—	33
Middlings	...	...	—	33
Maize (kibbled)	...	...	75	50-60
Maize germ-meal	...	} Contain very little vitamin B		
Maize gluten	...			
Buckwheat	...	...	80	70
Millet	...	...	55	50
Dari	...	...	60	50
Dried yeast	...	...	—	4
Baker's yeast	...	...	—	10-12
Marmite	...	...	—	8-10

As pointed out previously, some of the quantities for maintenance may be less than shown in this table. It is curious to see that the common cereal grains, wheat, rye, barley, have about the same value, that millet and dari have a rather greater value, maize a little less, and that oatmeal is least good in respect of vitamin B.

In general, our figures for maintenance are higher than those calculated from the data of Chick and Hume and of Cooper. The explanation is that our experiments were for a longer duration than 100 days. We have frequently seen polyneuritis on an amount corresponding to that of Chick and Hume after 15 weeks and occasionally after 20 or even 25 weeks. The minimum quantity adopted by us is that which keeps the birds in health for 26 or more weeks.

The relative value of the different cereals is shown better by adopting one of the above foods as a standard. It is most convenient to adopt the food richest in vitamin B, namely dried yeast, and give it the figure of 100. Then, by putting any of the above amounts for maintenance equal to 4 (dried yeast), the relative amount is given by calculating for 100 of the cereal.

Thus: 40 whole wheat = 4 dried yeast, therefore 100 whole wheat = 10;
50 whole wheat = 4 dried yeast, therefore 100 whole wheat = 8; 6 wheat germ = 4 dried yeast, therefore 100 wheat germ = 66.

The series of relative values is then as follows:

Relative vitamin B values of foods.

Dried yeast	100	Rye	9	Bran	12-13
Baker's yeast	33-40	Barley	7-8	Middlings	12-13
Marmite	40-50	Oats	4-5	Maize	7-8
Wheat germ	66	Oatmeal	4	Millet	8
Whole wheat	8-10	Buckwheat	5-6	Dari	8

It is further not difficult to calculate from these relative figures the quantity of food that must be contained in a diet. By dividing 400 by the above figures one obtains the percentage amount that should be included in a diet otherwise devoid of vitamin B. Another way of determining the amount of cereal, or a mixture of cereals, that should be present in a diet is by including the quantity, or quantities, that supply enough to give a total of 4. Thus, 20 % of millet will give 1.6, 10 % of dari will give 0.8, 40 % of whole wheat will give 3.2. Total = 5.6. The remainder of the diet can be white rice, or white flour, or fishmeal or any foodstuff without vitamin B.

These figures are for pigeons. For chickens, the amount must be calculated on a basis of 6, instead of 4. For rats, the vitamin B requirement is less, about half that of the pigeon, so that its amount will be calculated on a total of 2. The chief question is the requirement of man. He can be considered like a rat with the figure of 2, but it would be safer to consider that his requirement approaches that of a pigeon. The only figure that we can deduce for man is based on the experience in the Japanese Navy. Beri-beri was prevented by a ration of barley amounting to approximately one-third of the dry weight of the total food. This quantity, if it be assumed to be a minimum, gives a figure of not more than 3 for man's requirement, between that of a rat and a pigeon.

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