XCV. THE REARING OF CHICKENS ON THE INTENSIVE SYSTEM. PART III. B-VITAMIN REQUIREMENTS. COMPARISON OF YEAST EXTRACTS AND DRIED YEAST.

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IN our previous experiments [1922] it was shown that chickens could be reared from the day of hatching to maturity, under laboratory conditions, if the diet contained sufficient quantities of the three vitamins; if in addition, the protein in the diet were of "good" quality, the birds reached maturity at a rapid rate. The same diet has been used by Miss Malcolm and Mrs Pember on their poultry farm in Sussex and again the rapid growth to maturity with extraordinary health and vigour was observed.

In order to make the results of greater practical value and also to ascertain the real B-vitamin requirements, the ingredients of the diet needed simplification. As a substitute for caseinogen and secwa, dried skim milk is very suitable, since it contains the same substances, caseinogen, lactalbumin and lactose and probably also the same amount of B-vitamin. The replacement of oatmeal by other cereals presents a more difficult problem. Cereal grains contain B-vitamin in different and unknown quantities, so that the amount of marmite needed to balance the carbohydrate will vary for each one. It was originally supposed that the B-vitamin in oatmeal would be destroyed in the process of kilning used in its preparation, but this required verification; further all samples of oatmeal may not be prepared in the same way. Polished white rice forms the most convenient basal cereal, as it is free from B-vitamin. The amount of B-vitamin necessary to balance this food is likely to suffice for any other cereal. Chick and Hume [1917] have already found that 1 g. of yeast extract is a sufficient daily quantity to prevent polyneuritis in pigeons on a diet of white rice, but it does not follow that this quantity will be adequate for birds of other species. As will be shown later, the requirements of chickens for B-vitamin are greater than those of pigeons.

The determination of the total amount of B-vitamin, which is necessary, reckoned in terms of yeast extract, can only be made if all the constituents of the diet are free from this vitamin. Dried skim milk is almost certain to contain B-vitamin and must be substituted by a protein without it. Such a protein is egg-white. Very little B-vitamin is present in white fish as found by Cooper [1914]. White fish meal in small quantities could therefore be used instead of egg-white.

Marmite as the source of B-vitamin is too expensive for feeding animals. A cheaper form of yeast extract, cerema, has been introduced and dried yeast is another commercial article. The relative values of these varieties of yeast need determination. Marmite has been so frequently used in scientific investigations that it may be taken as the standard for comparison. These experiments have been designed to ascertain:

- (1) the relative values of marmite, cerema, and dried yeast as a source of B-vitamin;
- (2) the amount of marmite required to balance a diet of white rice and dried skim milk;
- (3) the amount of marmite needed to balance a diet of white rice and white fish meal;
- (4) by calculation from these data, the amounts of B-vitamin, in terms of marmite, in the foodstuffs used.

Complications could arise from the different amounts of salts, fat and A-vitamin in the diets. These foods contain very little fat, so that a constant supply of fat and A-vitamin could be introduced as cod-liver oil. The salt content of the diets are almost equal. No antiscorbutic was added because there are many observations that birds can exist for a considerable time without C-vitamin. The birds of these experiments have since been kept for over a year without C-vitamin, proving it is not needed (see Part IV, p. 787).

EXPERIMENTAL.

Housing. The housing provided for the chickens has consisted of ordinary commercial appliances. Two houses of each size were procured and placed side by side so that any comparisons, which were required, could be made under the same conditions. The houses were:

- (1) three compartment foster mother, or brooder, measuring $9' \times 2' 6'' \times 1' 6''$, one of the compartments being provided with an extra metal cover and heated by a paraffin lamp;
- (2) house measuring $7' \times 5' \times 5' 6''$ with run $4' \times 5' \times 3'$;
- (3) house measuring $6' \times 4' \times 4'$ with run $4' \times 4' \times 2' 6''$ and with three outside nest boxes fitted with trap nests;
- (4) house measuring $4' \times 3' \times 4'$ with temporary run $4' \times 4' \times 2' 6''$;
- (5) house measuring $6' \times 4' \times 4'$ with temporary run $5' \times 4' \times 2' 6''$;
- (6) fireless brooder measuring $1' 4'' \times 1' 4'' \times 1' 3''$. This was placed in a larger house (2) divided by wooden partitions into two equal parts.

The houses were provided with wooden floors and peat moss litter was used as covering.

No room was available for these houses inside the laboratory. They were placed upon the flat roof of the school building. No food other than that supplied could reach the birds, but in the autumn falling leaves from adjacent trees fell upon the roof and occasionally a few were blown into the runs. In our previous experiments it was found that housing in a room or upon a roof made no essential difference to the experimental results.

The houses provided for pairs of pigeons were double wire show cages for poultry, measuring $4' \times 2' \times 2' 3''$ protected at the sides by a metal piece and covered except at the front by wood. Each cage was fitted with a central perch.

Birds. The chicks, generally started as day-old, have been most kindly placed at our disposal by Miss Malcolm and Mrs Pember from their stock at Wellingham poultry farm, Lewes, and have consisted of Light Sussex except in one experiment in which a cross Wyandotte and Light Sussex was used.

Two pairs of pigeons were procured from a dealer.

Diet. The different diets and the results of the feeding are given under the several group headings numbered in continuation of our previous work. In general, the foodstuffs were mixed together as described in our earlier papers.

Comparison of the yeast extracts, marmite and cerema, on pigeons.

In testing foodstuffs for their B-vitamin value pigeons have been the experimental animal. Chick and Hume [1917] found that 1 g. of yeast extract per day was sufficient to prevent polyneuritis on a daily diet of 40 g. of white rice. In our former experiments with chickens 0.5 g. of marmite for 30 g. of oatmeal and milk and 0.25 g. for oatmeal with caseinogen and secwa in the diet was sufficient to prevent leg weakness. It appeared in the case of growing chickens that the carbohydrate of the food needed balancing by marmite. The balancing of the carbohydrate by B-vitamin has been considered necessary by former observers, but it is difficult to prove definitely.

As a preliminary to the comparison of marmite and cerema as a source of B-vitamin for growing chickens, an experiment was started on Feb. 7th, 1922, on two pairs of adult homer pigeons. One pair, red in colour, was fed upon white rice to which were added 0.75 g. of marmite for every 90 g. of rice and 0.75 cc. of cod-liver oil. The other pair, blue in colour, was given the same diet, but 0.75 g. of cerema instead of marmite. Neither pair of birds appeared to like the diet, so that after 14 days the cod-liver oil was omitted. On Feb. 24th, *i.e.* in the third week, both hens showed signs of leg weakness and a high stepping gait and were unable to get upon the perch; both cocks were also unwell. Their feathers were ruffled. Each bird was therefore dosed with 0.25 g. of marmite, or of cerema, dissolved in about 5 cc. of water on two consecutive days. This caused improvement in all the birds. The quantity of marmite, or of cerema, in the food was therefore not sufficient and was raised

on Feb. 27th to 1.5 g. per 90 g. of rice. This additional quantity did not, however, prevent the birds from getting ill again and they were all dosed with 0.5 g. of cerema, or of marmite, for three consecutive days. Their condition improved and their appetite increased. The marmite and cerema were both raised to 3 g. per 90 g. of rice on Mar. 7th.

A diet consisting only of polished rice and marmite, or cerema, cannot be considered adequate without additional protein, salts and A-vitamin. Fish meal (sample 1 see p. 785) was added on Mar. 11th.

On this diet, consisting of 90 g. white rice, 15 g. fish meal and 3 g. of marmite, the red pair of birds improved, gained in weight and the hen laid eggs on Apr. 5th. The blue pair of birds with 3 g. of cerema in the place of marmite did not show a similar improvement: their appetite fell off and they looked unwell with ruffled feathers. They were given a dose of 1 g. cerema on Mar. 27th and at the same time the amount of cerema in the food was raised to 3.75 g. per 90 g. of rice. This amount again did not suffice. The hen bird on Apr. 4th showed the characteristic symptoms of polyneuritis. She recovered after three doses of 1 g. cerema and was able to walk about the next day. On Apr. 12th the cerema was raised to 6 g., and on Apr. 23rd to 7.5 g. per 90 g. rice. The original supply of cerema was now exhausted. The new supply was quite different in appearance and it was learned that the original was from some old stock. The experiment was therefore stopped and both pairs of birds placed on oatmeal and fish meal to ascertain if C-vitamin was needed in their food (see Part IV, p. 787).

Thus, as far as marmite is concerned, 3 g. per 90 g. rice is sufficient to keep pigeons in health. The food consumption was on the average 45 g. rice and 7.5 g. fish meal per day. This is rather more than that given by Chick and Hume.

It may be thought that the salt addition with the fish meal was the reason for the maintenance of the red pair of pigeons, but this does not explain the failure of the blue pair on exactly the same diet.

With insufficient marmite or cerema not only did the appetite fall off, but also the weights. There was an increase of appetite after dosing and increase in weight. A steady increase in weight was shown by the red pair as soon as the balance of rice and marmite was reached. The weights are given in the following table:

	Red pair		Blue pair	
	Cock	Hen	Cock	Hen
	g.	g.	g.	g.
Feb. 9	445	439	434	327
" 15	435	430	429	318
" 22	409	391	392	286
Mar. 1	366	392	363	273
" 8	354	366	347	281
" 15	368	364	353	269
" 22	414	414	365	281
" 29	442	467	368	311
Apr. 7	438	450	407	321
" 19	447	472	398	284

GROUP IV. *Value of cerema and rice diet.*

Groups I to III of chickens had been kept on oatmeal and milk, or oatmeal and caseinogen and secwa with 0.75 g. of marmite per 90 g. of oatmeal and the results reported in our former papers [1922].

This experiment with Group IV was made to test the value of cerema as a source of B-vitamin for chickens. At the same time white rice was substituted for oatmeal and dried skim milk for caseinogen and secwa.

Twelve day-old Light Sussex chicks were used and the experiment was started on Feb. 22nd, 1922.

The diet was composed of equal parts of white rice and of dried skim milk. A-vitamin was added as cod-liver oil, at first 0.25 cc. but after the tenth day 0.5 cc. per bird per day. B-vitamin was included as cerema in the proportion of 0.75 g. per 90 g. of rice, the same quantity as the marmite used for oatmeal and milk. C-vitamin was not added as it was desired to see if chickens could be reared without it (see Part IV). During the second and third weeks the appetite of the birds was not good and it was not necessary to increase the daily food supply as often as expected. During the fourth week some slight signs of leg weakness were evident. On Mar. 30th at the beginning of the fifth week most of the birds showed very distinct signs of leg weakness. The amount of cerema in the food was then doubled. In addition, each bird was given a dose of 1 g. of cerema dissolved in about 5 cc. of water. This dosing was repeated on Apr. 4th and the amount of cerema in the food raised to 3 g. per 90 g. of rice. After a third dose the birds got slightly better, but still remained weak. Cerema was raised on Apr. 6th to 4.5 g. and on Apr. 11th to 6 g. per 90 g. of rice. The improvement was not maintained in spite of occasional doses of 1 g. of cerema. Ten birds died between Apr. 10th and May 19th. The remaining two recovered after raising the cerema to 7.5 g. per 90 g. of rice and were kept to maturity.

No definite result is given by the experiment, but it indicates that either more B-vitamin is required to balance rice than oatmeal, if cerema and marmite are equivalent, or cerema has less B-vitamin than marmite.

The record of the food consumption and weights of the birds is of no value.

GROUP V. *Value of cerema and oatmeal diet.*

The failure to rear the chickens of Group IV on equal parts of white rice and dried skim milk with the addition of 0.75 g. of cerema per 90 g. of rice led to a return to oatmeal as carbohydrate. Equal parts of oatmeal and dried skim milk give a food mixture of the composition:

Water	Ash	Fibre	Protein	Fat	Carbohydrate
8.1 %	4.7 %	0.5 %	21.9 %	4.3 %	60.6 %

with a protein to fat and carbohydrate ratio of 1 : 3.

This mixture corresponded fairly closely with the original mixture of

5/7 oatmeal, 1/7 caseinogen and 1/7 secwa used in the latter part of the experiment with Group III, namely

Water	Ash	Fibre	Protein	Fat	Carbohydrate
7.0 %	3.7 %	0.7 %	22.4 %	6.1 %	60.2 %

and protein to fat and carbohydrate ratio of 1 : 3.

Cod-liver oil was added to supply A-vitamin, at first at the rate of 0.5 cc., later 1.5 cc. per bird per day. Cerema was added to supply B-vitamin in the proportion of 0.75 g. per 90 g. of oatmeal. Orange juice was included for the first 37 days at the rate of 1 cc. per bird per day; it was afterwards omitted. Ten day-old Light Sussex chicks were used and the experiment started on May 4th, and concluded on Aug. 2nd, 1922.

Except for the accidental loss of five birds during the second week, owing to the paraffin lamp of the brooder smoking, the birds were raised to maturity without failure. The five dead birds were examined and no lesion could be discovered other than the lungs being filled with soot.

The food consumption and growth, owing to the accident, cannot be considered normal during the second and third weeks as all the birds were affected by the soot. The record is as follows:

		Oat-meal	Dried skim milk	Cod-liver oil	Cerema	Orange juice	Average weekly weights:					
		g.	g.	cc.	g.	cc.	g.	g.	g.			
May	5	30	30	2.5	0.25	10	May	4	39.4	July	1	806
"	6-29	60	60	5.0	0.5	10	"	10	59.2	"	12	1099
"	30-June 1	90	90	7.5	0.75	5	"	17	91	"	19	1282
June	2-9	120	120	7.5	1.0	5	"	24	155	"	27	1471
"	10-14	120	120	7.5	1.0	0	"	31	228	Aug.	2	1648
"	15	150	150	7.5	1.25	0	June	7	295			
"	16-26	180	180	7.5	1.5	0	"	14	421			
"	27-July 10	270	270	7.5	2.25	0	"	21	560			
July	11-Aug. 3	360	360	7.5	3.0	0	"	28	736			

No comparison of the rate of growth can be made with the birds of Group III which were white Leghorns.

On a carbohydrate basis of oatmeal 0.75 g. of cerema is sufficient per 90 g. This is the same as the quantity of marmite used in Group III. Cerema and marmite thus appear of equal value as a source of B-vitamin, if the amounts of B-vitamin in dried skim milk and in caseinogen and secwa are the same.

GROUP VI. *Comparison of marmite and cerema on chickens.*

The experiment with Group IV had shown that the addition of 0.75 g. of cerema per 90 g. of rice was not enough to maintain the birds. The experiment of Group V with 0.75 g. cerema per 90 g. of oatmeal indicated that this extract had an equal value to marmite. A thorough comparative experiment was now made on two lots of birds simultaneously, the one lot with cerema, as source of B-vitamin, and the other lot with marmite. The experiment with pigeons had shown that 3 g. of marmite per 90 g. of white rice was sufficient. This quantity of each extract was therefore included in the diet. Cod-liver oil to supply A-vitamin was added in the proportion of 0.5 cc. per bird per day. C-vitamin was omitted.

The diet was composed of equal parts of white rice and dried skim milk and had the following composition:

Water	Ash	Fibre	Protein	Fat	Carbohydrate
10.0 %	4.1 %	0.2 %	19.1 %	0.3 %	66.3 %

with a protein to fat and carbohydrate ratio of 1 : 3.5.

The experiment was started on June 26th, and concluded on Dec. 19th, 1922, at which date it was decided to put the hens and one cock into a single group on the diet with marmite for the purpose of procuring a second generation on a C-vitamin free food mixture (see Part IV, p. 787).

The constituents of the diet remained the same throughout; the only alteration was a reduction of the dried skim milk to 1/3 of the total on Aug. 21st (end of 8th week), mainly for the purpose of lessening the cost of feeding. The composition of the food mixture then became:

Water	Ash	Fibre	Protein	Fat	Carbohydrate
10.5 %	2.9 %	0.2 %	14.9 %	0.4 %	71.2 %

with a protein to fat and carbohydrate ratio of 1 : 4.8.

The group consisted of two lots of ten day-old chicks Wyandotte crossed with Light Sussex. Of these twenty birds, only two did not reach maturity. Both belonged to the cerema lot. One died on the 9th day of the experiment; no reason could be found on post mortem examination. The other was found paralysed in one leg at the end of the 9th week; post mortem examination indicated that this was due to pressure by a fibrous ovary on a nerve. On Sept. 28th, at the end of the 13th week, both groups showed signs of slight leg weakness. The birds were always wanting to rest and some of the cocks had a slight high stepping gait. Both the cerema and marmite were then raised, the marmite to 3.75 g. and the cerema to 4.5 g. per 90 g. of rice. The reason for the higher increase of cerema was because the birds of this lot seemed slightly worse. The symptoms disappeared in both lots in about 14 days.

Cocks of each lot started to crow during the 16th week.

On Nov. 11th (20th week) the birds of the cerema lot again showed slight signs of leg weakness. The cerema was then increased to 5.0 g. per 90 g. of rice. This figure was chosen so as to correspond with the nitrogen content of the preparations: marmite 5.7 %, cerema 4.3 %. After raising the cerema the symptoms disappeared.

On Nov. 15th (21st week) it became essential to separate the hens and cocks; the cerema lot became one cock and three hens, the marmite lot one cock and four hens. Excess cocks were kept in their separate lots. The hens of the cerema lot started laying on Dec. 10th (24th week), those of the marmite lot on Dec. 16th.

The total number of hens was too small to continue as two breeding pens. One cock and the seven hens were made into a single group and the experiment as regards comparison of cerema and marmite was terminated. The birds

were kept longer for more evidence about C-vitamin and its non-requirement by chickens (see Part IV, p. 787).

The two yeast extracts appear to be of nearly equal value; the two losses in the cerema lot cannot be fairly attributed to the presence of less B-vitamin. The early loss may have been due to a weaker chick at the start, the later one to some fault in sexual development. 5.0 g. of cerema is the equivalent of 3.75 g. of marmite per 90 g. of white rice with dried skim milk in the food. Throughout, the daily food consumption and the average weekly weights were very similar, as seen from the tables:

Daily food consumption.

	Rice g.	Dried milk g.	Cerema g.	Cod- liver oil cc.		Rice g.	Dried milk g.	Mar- mite g.	Cod- liver oil cc.
June 26	30	30	1.0	5	June 26	30	30	1.0	5
„ 27-July 7	60	60	2.0	5	„ 27-July 7	60	60	2.0	5
July 8-12	120	120	4.0	5	July 8-12	120	120	4.0	5
„ 13-20	150	150	5.0	5	„ 13-19	150	150	5.0	5
„ 21-23	180	180	6.0	5	„ 20	180	180	6.0	5
„ 24	240	240	8.0	5	„ 21-23	240	240	8.0	5
„ 25-Aug. 2	270	270	9.0	5	„ 24-31	270	270	9.0	5
Aug. 3-6	300	300	10.0	5	Aug. 1-2	300	300	10.0	5
„ 7-8	330	330	11.0	5	„ 3-8	330	330	11.0	5
„ 9-11	360	360	12.0	5	„ 9-11	360	360	12.0	5
„ 12-13	390	390	13.0	5	„ 12-13	390	390	13.0	5
„ 14-20	420	420	14.0	5	„ 14-16	420	420	14.0	5
„ 21-Sept. 6	570	285	19.0	5	„ 17-20	450	450	15.0	5
Sept. 7-25	780	390	26.0	5	„ 21-Sept. 6	600	300	20.0	5
„ 26-27	840	420	28.0	5	Sept. 7-25	810	405	27.0	5
„ 28-30	840	420	35.0	5	„ 26-27	870	435	29.0	5
Oct. 1-24	840	420	42.0	5	„ 28-Oct. 24	870	435	36.25	5
„ 25-31	900	450	45.0	5	Oct. 25-31	930	465	38.75	5
Nov. 1-10	960	480	48.0	5	Nov. 1-8	990	495	41.25	5
„ 11-Dec. 17	960	480	52.5	5	„ 9-Dec. 17	1050	525	43.75	5

Average weekly weights.

	Cerema lot g.	Marmite lot g.		Cerema lot g.	Marmite lot g.
June 26	40	40	Aug. 7	549	498
July 3	76	75	„ 10	628	579
„ 10	132	131	„ 21	845	797
„ 17	215	200	„ 28	988	972
„ 24	282	283	Sept. 4	1140	1123
„ 31	431	390	„ 11	1279	1307
			„ 25	1595	1653

	Cerema lot		Marmite lot	
	Cocks	Hens	Cocks	Hens
	g.	g.	g.	g.
Oct. 2	1927	1479	2062	1426
„ 9	2143	1594	2302	1572
„ 16	2351	1688	2486	1659
„ 23	2424	1713	2615	1700
„ 31	2701	1815	2876	1847
Nov. 8	2732	1872	2983	1931
„ 15	2862	1952	3032	1986
„ 22	2930	1985	3055	1993
Dec. 4	3070	2082	3198	2030
„ 11	3108	2225	3240	2090
„ 18	3192	2346	3288	2141

GROUP VII. *Comparison of cerema and dried yeast.*

Two lots of ten day-old Light Sussex chicks were used for this comparison, which was started on Dec. 22nd, 1922. The food for each lot consisted of two-thirds white rice and one-third dried skim milk. Cod-liver oil was added to each daily food supply at the same rate as before, 5 cc. per day, or 0.5 cc. per bird. C-vitamin was omitted. One lot received cerema in the proportion of 6 g. per 90 g. of rice. This quantity was chosen so as definitely to avoid any signs of leg weakness and was rather greater than the amount of 5 g. previously found to be sufficient. The other lot received 6 g. of dried yeast per 90 g. of rice. The two diets are not exactly equivalent on account of the higher nitrogen or protein content of dried yeast, 7.6 % against 4.3 % of cerema. This was unavoidable, as no similar protein could be added to compensate.

On Jan. 16th, 1923, the birds of the dried yeast lot showed distinct signs of leg weakness and drooping wings. The dried yeast was therefore raised to 12 g. per 90 g. of rice. On Apr. 6th, in the 15th week of the experiment, some signs of leg weakness became noticeable in the cerema lot; the amount of cerema was raised to 6.75 g. per 90 g. of rice. No other alteration was made and the experiment was terminated on Apr. 14th. Cocks of each group began to crow about Mar. 25th. The birds were mature and the hens needed separation from the cocks. The hens and a cock were continued as a single group for ascertaining egg production and hatchability on the C-vitamin-free diet (see Part IV). The first egg was laid on Apr. 29th in the 20th week.

Daily food consumption.

	Rice	Dried skim milk	Cerema	Cod-liver oil		Rice	Dried skim milk	Dried yeast	Cod-liver oil
	g.	g.	g.	cc.		g.	g.	g.	cc.
Dec. 22-27	60	30	4	5	Dec. 22-25	60	30	4	5
„ 28	90	45	6	5	„ 26-28	90	45	6	5
„ 29-Jan. 7	120	60	8	5	„ 29-Jan. 2	120	60	8	5
Jan. 8-9	150	75	10	5	Jan. 3-7	150	75	10	5
„ 10-11	180	90	12	5	„ 8-9	180	90	12	5
„ 12-14	210	105	14	5	„ 10-11	210	105	14	5
„ 15-19	240	120	16	5	„ 12-14	240	120	16	5
„ 20-24	270	135	18	5	„ 15-17	270	135	36	5
„ 25-29	300	150	20	5	„ 18-19	300	150	40	5
„ 30-31	330	165	22	5	„ 20-22	330	165	44	5
Feb. 1-7	360	180	24	5	„ 23-25	360	180	48	5
„ 8-9	360	195	26	5	„ 26-29	390	195	52	5
„ 10-13	420	210	28	5	„ 30-31	420	210	56	5
„ 14	450	225	30	5	Feb. 1-19	450	225	60	5
„ 15-19	480	240	32	5	„ 20	480	240	64	5
„ 20	510	255	34	5	„ 21-23	540	270	72	5
„ 21-23	540	270	36	5	„ 24-26	600	300	80	5
„ 24-26	600	300	40	5	„ 27-Mar. 6	660	330	88	5
„ 27-Mar. 6	660	330	40	5	Mar. 7-30	720	360	96	5
Mar. 7-Apr. 5	720	360	48	5	„ 31-Apr. 13	780	390	104	5
Apr. 6-13	720	360	54	5					

Altogether three birds out of 20 were lost. One bird of the cerema lot died on the second day of the experiment. Another bird of this lot dislocated

its leg in the second week and was chloroformed. The third bird belonged to the dried yeast group and was found dead on Feb. 6th in the 7th week; it had suffered more severely than the other birds from insufficiency of dried yeast before the quantity was raised and owing to its weakness may have been crushed by the birds huddling together at night. Excluding the accidental loss, 90 % of each lot was raised to maturity. Just before the conclusion of the experiment, on Apr. 7th, a cock of the cerema lot was found with a damaged leg, evidently from fighting and was chloroformed.

The close parallelism between the two lots is shown by the daily food consumption (p. 780) and the weekly average weights of the birds which were:

	Cerema lot	Dried yeast lot		Cerema lot	Dried yeast lot
	g.	g.		g.	g.
Dec. 22	37.6	36.7	Feb. 23	921	992
" 29	76.6	74.8	Mar. 2	1072	1089
Jan. 5	132.4	137.6	" 9	1262	1334
" 12	220	234	" 16	1418	1508
" 19	284	309	" 23	1596	1598
" 26	386	400	" 30	1801	1840
Feb. 2	514	502	Apr. 6	1920	2052
" 9	604	665	" 13	2034	2144
" 16	748	810			

The higher consumption in the dried yeast lot was due to its having one or two more birds at certain periods, as the result of losses in the cerema lot. The food consumption per bird is approximately the same in both lots.

The advantage, if any, is on the side of the dried yeast lot. The two lots contained the same number of cocks and hens.

It is interesting to note that these weights run very close to the weights of the birds in Group VI. Group VI was started in June, Group VII in December. There is no appreciable difference in the rate of growth at different seasons of the year, as is commonly believed by poultry keepers. The result shows that a proportion of 12 g. of dried yeast is equal to 6, or more probably, 6.75 g. of cerema per 90 g. rice on a diet of two-thirds white rice and one-third dried skim milk.

GROUPS VIII and IX. *Amount of marmite required for a diet containing no B-vitamin.*

In order to determine the full amount of marmite which must be included in a diet not containing B-vitamin in any of its ingredients, it was necessary to replace the dried skim milk in the diet of the previous groups by some protein with little or no B-vitamin. Cooper [1914] has shown that more than 10 g. per day of white fish was required to prevent polyneuritis in pigeons. The total amount necessary was not determined, but this result suggests that white fish flesh has very little B-vitamin. Dried commercial white fish meal thus seemed a suitable protein for these experiments, if used in small amounts. 5 g. of fish meal with 90 g. of rice would not be likely to introduce an appreciable amount of B-vitamin.

Fish meal contains a large proportion of mineral matter, so that a further ash supplement is unnecessary. Some samples of fish meal have a low fat content (see p. 785 for analyses) and the amount of A-vitamin in this fat is probably small. Fish meals, dark in colour, are evidently made at a high temperature and some, if not all, of the A-vitamin would be destroyed. A known amount of fat supplying A-vitamin in the form of cod-liver oil was therefore added.

The quantity of marmite to be included as source of B-vitamin required consideration. Group VII showed that 6 g. of cerema or 12 g. of dried yeast was adequate for 90 g. of rice and 30 g. of dried skim milk. This amount was likely to be too small on the fish meal diet, since the dried skim milk probably carried enough B-vitamin to balance its carbohydrate, fat and protein. The dried skim milk supplied 28 parts of combustible food material and the rice supplied 58 parts. Half as much cerema again would be required to balance the 28 parts derived from the dried skim milk, now replaced by fish meal and a proportion of rice. Consequently for the fish meal diet containing 90 g. of rice and 5 g. of fish meal $6 + 3 = 9$ g. of cerema would be needed. Cerema and marmite are nearly equal, so 9 g. of marmite was added to balance these amounts and a small allowance of cod-liver oil, 0.5 cc. per 90 g. of rice.

The diet composed of 90 g. of rice, 5 g. of fish meal (sample 2), 0.5 cc. of cod-liver oil and 9 g. of marmite had the composition:

Water	Salt	Ash	Fibre	Protein	Fat	Carbohydrate
12.7 %	1.7 %	2.7 %	0.3 %	11.3 %	1.0 %	70.6 %

with a protein to fat and carbohydrate ratio of 1 : 6.3. Marmite is present in the proportion of 8.6 %.

In order to ascertain if the amount of marmite was properly adjusted, the cod-liver oil was raised at intervals to 1.0, 1.5, 2.0, 3.0, 4.0, 5.0 cc. At the same time the effect of the extra cod-liver oil and its relation to the amount of B-vitamin would be shown. Our previous experiments (Part I) had indicated that extra cod-liver oil needed balancing with B-vitamin. This question is being further studied in another group of birds.

GROUP VIII consisted of 11 Light Sussex birds, 7 weeks old, previously kept on a rice and dried skim milk diet as used for Group VII. The experiment was started on Mar. 9th and concluded on June 21st, 1923 (14 weeks). The birds did not grow rapidly and there was considerable difference in the weights of the birds of the same age and sex, particularly noticeable after the oil was raised to 2.5 and 4 cc. Some of the hens became adult long before others. One of them had a sort of spinal curvature. The birds were not active and always wanted to rest. Signs of leg weakness with high stepping gait and drooping wings were observed. Their feathering was slow and their combs were pale. The cocks were never heard to crow. All the birds were raised to maturity. The general appearance of the birds was the best indication that the diet contained just the sufficient amount of B-vitamin to maintain them and take them to maturity, but not enough to keep them in

good health. The conditions of the experiment were repeated in the next group.

GROUP IX consisted of 10 Light Sussex day-old chicks, which were fed on the same diet as Group VIII. The experiment was started on Apr. 7th and concluded on Aug. 24th, 1923 (20 weeks).

Between May 4th and June 19th, the cod-liver oil was gradually raised from 0.5 cc. to 4 cc. (see table of weights). With the increase of the cod-liver oil the birds had the appearances of those of Group VIII, pale combs, slight leg weakness, high stepping gait, poor feathering, which pointed to an insufficiency of B-vitamin in the diet. The marmite was therefore raised on June 30th (13th week) to 9.75 g. per 90 g. rice. Some improvement followed. It was raised again on Aug. 3rd (17th week) to 10.5 g. per 90 g. of rice. The general improvement was more noticeable.

The food intake was not measured daily as in other groups. The food mixture was made up in sufficient quantity to last for two or three days. The birds were allowed as much as they would eat; food was constantly before them. The actual consumption has no particular bearing on the observations.

Three birds were lost during the experiment. One was unwell on arrival and died the first day. The second died on June 20th. This bird had the most severe symptoms of leg weakness in the group. Post mortem examination showed considerable abnormality of the gut, the walls of which were very thick. The third died on Aug. 7th. This bird had stoppage and distention of the gut at the junction of the caeca. The connective tissue holding the intestines was very tight and fibrous.

One of the birds raised to maturity showed some deformity in one wing, probably from resting on it during the time the leg weakness was observed.

It is not possible to compare the growth with that of the birds of Groups VI and VII, which had not only more protein, but also milk protein in the diet. In order to see the irregularity the weekly weight record of each bird must be recorded (see p. 784).

The weekly weights are not the best indication of the condition of the birds. The condition can really only be judged by comparison with other birds which have been reared on a diet containing all the constituents in better proportions. At 20 weeks the birds were not mature. The cocks never crowed and the hens were small for their age. It was difficult to be certain of the sex of two of the birds, Nos. 2 and 6. Only at the end did their sex become quite certain.

The results confirm those of Group VIII. On a diet of 90 g. rice, 5 g. fish meal and 0.5 cc. cod-liver oil, 9 g. of marmite is sufficient to maintain and raise them to maturity. Excess of cod-liver oil produced signs of leg weakness. These observations do not agree with those of Emmett and Peacock [1923] who attribute leg weakness and drooping wings to lack of A-vitamin. A large excess of cod-liver oil was present in the diets of Groups VIII and IX with scarcely enough marmite to balance the rice and fish meal adequately. Our

result suggests that the diet used by Emmett and Peacock had not enough B-vitamin in it. Perfect health and growth depend upon a proper amount of all the constituents in the diet. The question of the balance of carbohydrate, fat and protein with B-vitamin is now under investigation.

Average weekly weights, and time of increase of cod-liver oil and marmite.

	Apr. 7	Apr. 13	Apr. 20	Apr. 27	May 4	May 11	May 18	Sex
Cod-liver oil	0.5 cc.	0.5 cc.	0.5 cc.	0.5 cc.	1 cc.	1 cc.	2 cc.	
Marmite	9.0 g.	9.0 g.	9.0 g.	9.0 g.	9.0 g.	9.0 g.	9.0 g.	
No. 1	41.5 g.	59.5 g.	95.0 g.	150 g.	205.0 g.	303.0 g.	385 g.	Cock
" 2	41.0	66.0	82.0	123	182.0	230.0	317	Cock
" 3	41.0	62.5	83.0	125	152.0	221.0	293	Cock
" 4	42.5	63.5	84.0	129	173.0	242.0	329	Hen
" 5	39.0	61.5	82.0	117	157.0	219.0	292	Hen
" 6	38.5	59.5	84.0	115	149.0	202.0	250	Cock
" 7	40.0	53.5	64.0	88	111.0	159.0	203	Hen
" 8	40.5	63.5	81.0	109	139.0	190.0	238	
" 9	38.5	49.5	58.0	80	97.0	134.0	184	
" 10	41.0	Died						
Average	40.3	59.7	79.9	115	151.5	211.0	277	
Increase	—	19.4	20.2	35	36.5	59.5	66	

	May 25	June 1	June 8	June 15	June 22	June 29	July 6	Sex
Cod-liver oil	2.5 cc.	2.5 cc.	2.7 cc.	3.5 cc.	4.0 cc.	4.0 cc.	4.0 cc.	
Marmite	9.0 g.	9.0 g.	9.0 g.	9.0 g.	9.0 g.	9.75 g.	9.75 g.	
No. 1	519 g.	580 g.	800 g.	905 g.	1140 g.	1325 g.	1526 g.	Cock
" 2	398	520	682	802	935	1100	1241	Cock
" 3	393	555	675	823	975	1165	1314	Cock
" 4	429	525	635	777	957	1105	1257	Hen
" 5	365	495	605	690	840	915	1055	Hen
" 6	334	425	505	603	680	765	863	Cock
" 7	253	330	352	422	488	555	612	Hen
" 8	312	417	498	568	665	775	854	
" 9	234	300	402	517	Died			
Average	360	494	573	678	835	963	1080	
Increase	83	134	79	105	157	128	117	

	July 13	July 20	July 27	Aug. 3	Aug. 10	Aug. 17	Aug. 24	Sex
Cod-liver oil	4.0 cc.	4.0 cc.	4.0 cc.	4.0 cc.	4.0 cc.	4.0 cc.	4.0 cc.	
Marmite	9.75 g.	9.75 g.	9.75 g.	10.5 g.	10.5 g.	10.5 g.	10.5 g.	
No. 1	1640 g.	1900 g.	1940 g.	2170 g.	2250 g.	2450 g.	—	Cock
" 2	1312	1480	1605	1730	1940	2100	—	Cock
" 3	1410	1515	1600	1800	1950	2025	—	Cock
" 4	1207	1400	1560	1725	1890	1960	—	Hen
" 5	1127	1198	1280	1420	1570	1675	—	Hen
" 6	920	1012	1122	1235	1325	1480	—	Cock
" 7	659	700	725	800	990	995	—	Hen
" 8	922	919	857	905	Died			
Average	1149	1265	1336	1473	1702	1812		
Increase	69	118	71	137	229	110		

The comparative B-vitamin value of foodstuffs.

The comparative value of several foodstuffs in preventing polyneuritis in pigeons has been determined by Chick and Hume [1917] and Cooper [1914]. Yeast extract (marmite) was found to contain more B-vitamin than any of the others except rice germ. The B-vitamin content of marmite has also been found to be very constant. The constancy has again been shown by our experiments with pigeons (p. 774). For purposes of comparison, marmite may be chosen as the most convenient standard. If the figure of 1 g. found by

the above observers be put at 100, their figures give the following comparative numbers:

Marmite	100	Ox liver	33	Ox heart muscle	20
Wheat germ	67	Barley, unhusked	27	Ox brain	17
Pressed yeast	40	Barley, husked	20	Sheep brain	8
Lentils	33	Peas	20	Beef muscle	5
Egg yolk	33				

Groups VI and VII showed that 1.25 g. marmite were equal to 1.66 g. of cerema and 2 to 2.25 g. of cerema were equal to 4 g. of dried yeast. Hence, if marmite = 100, cerema = 75, dried yeast = 50.

Groups VIII and IX were raised to maturity on the diet of 90 g. of rice and 5 g. of fish meal with the addition of 9 g. of marmite, or 8.6 % of the total food.

Group VI, on a diet of equal parts of rice and dried skim milk, needed an addition of 3 g. of marmite per 90 g. of rice. Assuming that 90 g. of dried skim milk require 9 g. of marmite, as only 3 g. were wanted, this quantity of dried skim milk would contain the equivalent of 6 g. of marmite, or 6.7 %.

It is not possible to calculate from the diet of Group V, equal parts of oatmeal and dried skim milk, the B-vitamin value of oatmeal; but in a further experiment (Group XI, see Part V) chicks were raised to maturity on a diet of 90 g. of oatmeal and 5 g. of fish meal with an addition of 1.5 g. of marmite. The 95 g. of food would thus contain an equivalent of $9 - 1.5 = 7.5$ g. of marmite or 7.9 %.

Oatmeal, as a cereal, thus contains a considerable amount of B-vitamin, but it is not sufficient for the rearing of chicks. It does not follow that whole oats contain the same amount, nor that other cereals and cereal products have an equal amount. Germ meals may contain more. Further feeding experiments will be necessary to ascertain the amounts. In the meanwhile, in order to be on the safe side for maintaining the health of chicks, the addition of some foodstuff rich in B-vitamin is advisable. The maximum needed would be 8.6 % in terms of marmite or 17 % in terms of dried yeast. With a cereal such as oatmeal an addition of 1.5 % of marmite or 3 % of dried yeast would be adequate.

Analyses of foods.

The foods which have been used in these experiments had the following composition, from which the composition of the actual diets have been calculated:

	Water %	Salt %	Other ash %	Fibre %	Protein %	Fat %	Carbohydrate	
							By diff. %	Undet. %
Oatmeal*	7.5	—	1.9	1.0	12.0	8.4	69.4	—
Rice white*	11.4	—	0.6	0.3	6.5	0.4	80.8	—
Dried skim milk (1)	7.0	—	7.6	—	31.8	0.1	43.3†	10.2
" " (2)	8.6	—	7.6	—	31.8	0.2	43.4†	8.4
Fish meal (1)	14.7	3.1	18.6	—	60.5	3.3	—	—
" " (2)	12.1	3.2	21.3	—	59.8	1.9	—	—
Marmite	27.3	14.5	13.9	—	32.5	—	11.8	—
Cerema	28.6	6.9	10.5	—	24.4	—	29.6	—
Dried yeast	2.5	—	8.6	—	43.5	0.9	44.5	—

* From *Analyses and Energy Values of Foods* by R. H. A. Plimmer.

† Lactose by analysis.

SUMMARY.

1. Pigeons on a diet of polished rice (90 g.) and fish meal (15 g.) need an addition of 3 g. of marmite for maintenance.
2. Chicks on a diet of equal parts of white rice and dried skim milk need an addition of 3 g. of marmite per 90 g. of rice, or 3·3 %.
3. Chicks on a diet of equal parts of oatmeal and dried skim milk need an addition of 1·5 g. of marmite per 90 g. of oatmeal, or 1·6 %.
4. Chicks on a diet of 90 g. of rice and 5 g. of fish meal with 0·5 cc. of cod-liver oil need an addition of 9 g. of marmite, or 8·6 % of the food.
5. If marmite be put at 100, cerema has a value of 75, dried yeast of 50, oatmeal of 7·9, dried skim milk of 6·7.

We gratefully acknowledge supplies of marmite, cerema and dried yeast from the Marmite Food Extract Co., Ltd.

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