MAY 1945 RESEARCH BULLETIN 390

UNIVERSITY OF MISSOURI COLLEGE OF AGRICULTURE
AGRICULTURAL EXPERIMENT STATION

M. F. MILLER, *Director*

Critical Vitamins in Rations for Baby Chicks

L. R. RICHARDSON, A. G. HOGAN and H. L. KEMPSTER

(Publication Authorized May 10, 1945)

COLUMBIA, MISSOURI

SUMMARY

The basal ration was adequate in protein, minerals, and energy content. It contained no water-soluble vitamins except those supplied by 22.2 per cent of yellow corn, 3 per cent of wheat bran, 9 per cent of wheat middlings, and 15 per cent of soybean meal. It was grossly inadequate for the chick and presumably it was as deficient in the vitamins of the B-complex as any ration that would ever be encountered in practice. When the recognized members of the vitamin-B complex were added to the basal ration it supported a normal rate of growth. Each of these vitamins was omitted separately from the mixture in order to determine which were present in the basal diet in insufficient quantity.

Thiamine and pyridoxine were present in adequate amounts.

When pantothenic acid was omitted from the vitamin mixture the chicks developed symptoms similar to those frequently seen in chicks sent in by poultrymen for examination because of an excessive rate of mortality. A deficiency of pantothenic acid is probably an important cause of subnormal vitality in baby chicks.

The basal diet is deficient in choline. If the constituents of a ration are properly selected this nutrient should be present in adequate amount, but in practice mild deficiencies of this vitamin probably do occur.

If nicotinic acid was omitted from the vitamin mixture the rate of growth was depressed and the incidence of perosis was increased. It may be that nicotinic acid is never deficient in a practical ration but the possibility deserves further study.

There was a mild deficiency of biotin but it is doubtful that there would ever be a deficiency in a practical ration.

When all recognized vitamins were added to the basal diet about 2 per cent of the chicks still developed perosis. It was concluded that this was due to a mild deficiency of an unrecognized vitamin.

It was suggested that it may become practical in the future to fortify the rations of baby chicks with synthetic vitamins.

Critical Vitamins in Rations for Baby Chicks

L. R. Richardson, A. G. Hogan and H. L. Kempster

On various occasions farmers have reported to the Missouri Agricultural Experiment Station that the mortality rate in their baby chicks, or turkey poults, was excessively high. Examination by the Station pathologist usually failed to identify any infectious disease, and it is quite certain that heavy losses in this locality during the first three or four weeks after hatching are frequently due to other causes. It can be taken for granted that an unsuitable environment accounts for some of the losses. However, a preliminary investigation indicated that malnutrition, due chiefly to vitamin deficiencies, bears part of the responsibility, and an attempt was made to discover which of these nutrients are most likely to be deficient.

EXPERIMENTAL

The chicks were male Barred Plymouth Rocks, and they were quartered in rooms which were maintained at an even temperature by automatic heat controls. The batteries in which the chicks were confined the first three weeks had electric heating units, connected with thermostats, which kept the inside temperature between 90° and 95°F.. At the beginning of the fourth week the chicks were transferred to another room in which the temperature was kept between 80° and 85°F. Water and the experimental diets were constantly available after the chicks were first transferred to the batteries. Usually there were from 5 to 7 chicks on each diet, but each feeding trial was repeated five or six times. So far as was possible each group contained chicks of equal weight. None was rejected unless it was obviously inferior, and all mortalities were counted after the first week. There will be some mortalities after the first week on any ration when these practices are followed, but in our opinion this procedure accentuates the difference between a superior and a mediocre ration. This is the point of chief interest to us at present.

A few attempts have been made to determine which vitamins were deficient in feeds that were brought in from the field by poultry growers reporting them to be unsatisfactory. However, the feeds obtained from these sources were not satisfactory for the purpose. The response of the chicks which received them was highly variable, which indicated that the feeds were border-line in adequacy. In order to identify the specific vitamins which were lacking under these circumstances, many more chicks would be required than our facilities would accommodate,

and the method of attack was changed. It was decided to begin with a ration that was made up of the most commonly used feedstuffs, and that contained none of the more reliable vitamin carriers such as dried skim milk or alfalfa meal. It had been shown by actual trial that this ration gave reasonably satisfactory, though less than optimum, results. In order to reduce the vitamin content of this diet to a lower level than one would ever expect a practical poultryman to employ in the field, the practical diet was diluted almost 50 per cent with a synthetic diet which contained no added vitamins. The composition of the modified diet, Ration A, is shown in Table 1.

TABLE 1.—COMPOSITION OF EXPERIMENTAL DIET A

	I		II
Yellow corn	22.2	casein	20.0
Wheat bran	3.0	gelatin	5.5
Wheat middlings	9.0	glucose	14.8
Soybean meal	15.0	cellulose	1.2
Steamed bone meal	1.8	lard	5.2
Iodized salt	0.3	mineral salts	2.0
	51.3		48.7

Manganese, 12 mg.	per 100 grams
Vitamin A, 2000 I. U.	" " "
Vitamin D, 283 I. U.	" " "
Alpha tocopherol, 2.5 mg.	" " "
2-methyl-1, 4-naphthoquinone, 2.5 mg.	" " "

Ration A is at least reasonably adequate in protein, energy, and minerals, and it is markedly deficient in certain of the water-soluble vitamins. Our object was to identify the recognized vitamins that are deficient, and to determine whether any of the unrecognized vitamins are also seriously deficient. The procedure was to add all of the vitamins to Ration A that one could suppose might be present in insufficient quantity, and then to omit each one separately from the mixture. The amounts of the added vitamins, and the response of the chicks are shown in Table 2. In a few instances the quantities of the vitamins supplied were not exactly the same as is indicated in the table, but since the changes had no apparent significance they will not be described.

The statistical analysis of the differences in weight is shown in Table 3. There was a considerable number of preliminary feeding tests in which only a few rations were tried at one time. All of the chicks are included in Table 2, but only those under observation at the same time, and from the same shipments, are compared in Table 3. This explains why the number of chicks on the various rations, as shown in Tables 2 and 3 is not the same.

The first point to emphasize is the excellent rate of growth of the chicks in Groups 1, 2, and 4. It will be recalled that Ration A was

TABLE 2. - Vitamin deficiencies of Ration A

Group	No. of chicks	Added vitamins,[1] per 100 gms.							Record at 4 weeks of age			
		THIAMINE	RIBO FLAVIN	PYRIDOXINE	CA-PANTOTHENATE	NICOTINIC ACID	CHOLINE	BIOTIN	Weight	Perosis		Mortality
		mg.	mg.	mg.	mg.	mg.	mg.	mcg.	gm.	% [2]	Score[3]	%[4]
1	101	0.4	0.4	0.4	2	5	400	20	301	4.0	70	2.0
2	35		0.4	0.4	2	5	400	20	298	3.0	50	5.8
3	4	0.4		0.4	2	5	400	20	129	75% curled toe paralysis		
4	53	0.4	0.4		2	5	400	20	293	1.9	75	1.9
5	37	0.4	0.4	0.4		5	400	20	204	27.	50	10.8
6	44	0.4	0.4	0.4	2		400	20	268	28.	67	2.3
7	35	0.4	0.4	0.4	2	5		20	238	67.	56	8.5
8	84	0.4	0.4	0.4	2	5	400		284	11.	61	4.8
9	5		0.4		2	5	400	20	317	0	--	0

1. Supplied by Merck and Co., Rahway, N. J.
2. Percent of chicks that survived.
3. One + indicates definite perosis, 4 + indicates the highest degree of severity. In calculating the average numerical score 1+ = 25, 3+ = 75, and 4+ = 100. The average numerical score is based on the number of affected chicks.
4. Chicks that died during the first week are not included in the computation.

TABLE 3. - Significance of Differences in Weight[1]

No. of Chicks	Group and ration	F Ratio [2]		Mean	σ_m	Difference of Means	σ_D
		Obtained	of .01 Point [3]				
37	1 control	60.92	7.12	312.1	8.63	98.8	14.06
28	5 No Pantothenic Acid			213.3	11.46		
44	1 control	22.86	7.04	311.2	11.88	71.2	15.91
31	7 No Choline			240.0	8.61		
58	1 control	9.83	6.90	306.9	11.84	39.8	14.11
53	8 No Biotin			267.1	7.06		
44	1 control	8.52	7.01	311.2	11.88	42.8	15.34
43	6 No Nicotinic Acid			268.4	9.64		

1. This table was prepared by Prof. B. H. Frame of the Department of Agricultural Economics.
2. Statistical Methods, by George W. Snedecor, Iowa State Agricultural College (1)
3. Any ratio higher than is shown below indicates a probability less than .01 that the difference is due to chance.

purposely made lower in vitamin content than any poultry grower is ever likely to employ in practice. However, when this ration was supplemented with seven, or even five, water-soluble vitamins the chicks grew more rapidly than would be expected from current growth standards. This indicates that none of the unrecognized vitamins is likely to be seriously deficient in a practical chick starter ration. In other words, when a practical ration is deficient in vitamins, the deficiencies can be remedied by providing vitamins that are now recognized. When reliable assay methods for these vitamins become available it should be possible to determine whether or not a ration contains an adequate vitamin supply without a feeding trial.

It should be pointed out that the rapid rate of growth on the supplemented ration is due partly to the inclusion of casein and gelatin. In our experience the rate of growth on rations that contain these two proteins is higher than when they are replaced by the commonly used protein supplements.

Thiamine and Pyridoxine. It can be taken for granted, with almost complete certainty, that failures with a chick starter ration are not due to a deficiency of either thiamine or pyridoxine. When either vitamin was omitted or when both were omitted, the chicks grew at the same rate as when they were included.

Riboflavin. It had been demonstrated previously, in trials that will be described in a separate publication, that rations can be much better than Ration A and still be deficient in riboflavin. For that reason only one trial, with 4 chicks, was conducted with a ration that did not contain added riboflavin. There were no deaths within the 4-week period, but growth was very slow and 3 chicks developed typical curled-toe paralysis. It is well known that practical rations will be deficient in this vitamin unless it is provided in such supplements as alfalfa meal or milk, or milk by-products. Since this vitamin is labile, the quality of the supplements is of decisive importance.

Pantothenic Acid. Ration A is seriously deficient in pantothenic acid. The weights were subnormal, many of the chicks had a characteristic staggering gait, and they were markedly listless. Of the 33 chicks that survived, 4 had severe perosis. There were 5 cases marked as mild, but there may have been some confusion between mild perosis and incoordination of the muscles. None developed dermatitis, but the mortality rate was higher than is normal. The listless attitude, and posture, were strikingly similar to those exhibited by many of the chicks that had been brought to us by practical poultry growers for examination because of an abnormally high mortality rate. This similarity in appearance indicates to us that at the present time a deficiency of pantothenic acid is the most important single cause of death in young chicks, when the management is reasonably satisfactory in other respects. As yet, however, there has been no opportunity to test this hypothesis.

Nicotinic Acid. Although it had been demonstrated by Briggs et al. (2a, b) that the chick requires nicotinic acid, it was not expected that Ration A would be deficient in that vitamin. However, the weights were slightly, but significantly, reduced when nicotinic acid was omitted from the diet. The most striking result of the omission, however, was the more frequent occurrence of perosis, which was nearly 24 per cent. It is quite certain that under some circumstances a deficiency of this vitamin will increase the incidence of this abnormality, but it may be that the prevention of perosis is not a specific function of nicotinic acid. One could assume that the absence of nicotinic acid depresses the rate at which intestinal bacteria synthesize a perosis-preventing vitamin. It is doubtful that a practical ration would ever be sufficiently deficient in nicotinic acid to depress the growth rate. However, the possibility that a mild deficiency may increase the incidence of perosis is worthy of consideration.

Choline. The subnormal weights, together with the high incidence of perosis, show very clearly that Ration A is seriously deficient in choline. If Fraction II of Ration A were replaced with an equal weight of Fraction I, the amount of choline in the modified diet would be almost doubled, and this quantity might be enough to prevent perosis. It is quite certain though that this quantity of choline would not be enough to provide a wide margin of safety. Perosis occurs frequently in farm flocks in this section of Missouri and in our opinion a deficiency of choline is a contributing factor.

Biotin. The weights of chicks which did not receive biotin were slightly below the maximum, and the incidence of perosis was slightly higher than when this vitamin was supplied. The statistical analysis in Table 3 was based on six separate comparisons, with from 4 to 15 chicks in each group that did not receive biotin. The difference between the controls and experimental groups is statistically significant. There were 27 survivors in Group 8 of Table 2 that do not appear in Table 3, and it happened that the average weight of these 27 chicks was higher than the average of the 53 that were available for statistical analysis. This explains why the difference in weight between Groups 1 and 8 is less in Table 2 than it is in Table 3. It seems quite certain, however, that Ration A is mildly deficient in biotin, though it seems improbable that a practical ration would be deficient in this vitamin.

DISCUSSION

On several occasions local poultry growers have reported an excessive mortality rate in baby chicks. No extensive investigations have been carried out to determine the specific cause of the deaths but the case histories indicated that mildly deficient rations and an unsuitable environment bore the chief responsibility. A combination of these two

factors is usually the most probable explanation. It was our opinion in a few cases we investigated that the chicks were too cold, at least during certain intervals, and that the rations were somewhat less than adequate. If there had been only one unfavorable factor the losses would have been light, but when there were two the losses were heavy. Data to support this hypothesis will be described in detail in another publication. One group of chicks was subjected to a temperature that was uncomfortably cool, and another was supplied a ration that was deficient in a vitamin, with little or no increase in the mortality rate. However, when these unfavorable conditions were imposed simultaneously the mortality rate was disastrous. This suggests that when a ration is to be tested for adequacy, or when the optimum quantity of a vitamin is to be determined, the resistance to low temperatures, or to a sudden drop in temperature, is as important as the rate of growth.

It is clear from the data we have presented that Ration A is seriously deficient in several recognized vitamins and it is our opinion that under practical conditions, when there are no lapses in management, some of these deficiencies are an important cause of early death in baby chicks. Ordinarily these deficiencies are prevented by including in the diet the more available vitamin carriers such as alfalfa meal, dried skim milk, or a miscellaneous group of factory by-products. If these supplements are seriously defective, or if they are included in insufficient quantity, the ration will be deficient in essential vitamins.

As to which of the vitamins may be limiting factors in the field we are unable to say. Most of the practical rations we prepared in the laboratory contained 25 per cent or more of soybean meal, and in our experience this quantity provides a high degree of protection against vitamin deficiencies. These rations were deficient in riboflavin but when this vitamin was supplied there was never a clear indication that any other recognized vitamin was a first limiting factor. However, if other protein supplements replace most of the soybean meal one could not take it for granted that other members of the B-complex are always present in sufficient quantity. In fact experience shows that some of them are at times present in insufficient quantity. The data reported in Table 2, and observations on chicks brought in from the field, convince us that deficiencies of pantothenic acid are almost as serious as are deficiencies of riboflavin. The symptoms in chicks sent in for examination are usually similar to those we observed in Group 5, which received an insufficient amount of pantothenic acid. In all probability it would be possible to make up practical rations that are deficient in nicotinic acid and choline, but we have no definite evidence on the frequency with which such deficiencies occur. Biotin is only slightly deficient in Ration A, and it is probably present in adequate amount in any practical ration. The data in Table 2 suggest that the adequacy of practical rations could be insured by adding synthetic

vitamins to them but our experience is not as yet sufficiently extensive to permit a final decision.

Although Ration A is deficient in recognized vitamins there is no reason to suppose it is seriously deficient in any unrecognized vitamin. There is reason, however, to suspect that it is mildly deficient in an unknown factor. It will be noted that in Table 2 only Group 9, which contained 5 chicks, was entirely free from perosis. Groups 1, 2, and 4 presumably received an adequate supply of all known vitamins, but this abnormality occurred in every group. It is our opinion that these cases are due to a slight deficiency of an unrecognized vitamin, and that they never occur when the ration is entirely adequate. Hill and coworkers (3) concluded that certain chick rations developed to meet war-time conditions are deficient in one or more unidentified vitamins but this conclusion was apparently based on the calculated vitamin content of their experimental diet. However, it is our experience that even when a ration is adequate by all conventional standards the rate of growth may be greatly accelerated by the addition of certain crude vitamin carriers. In our opinion this is due to the addition of vitamins that have not as yet been recognized.

At the time the feeding trials just described were under way several suggestions came to us which we plan to investigate with some care. There has been no opportunity as yet to establish their merit but they seem sufficiently important to justify some mention.

Repletion Rations. Our experience has led us to a working hypothesis that the mortality rate in baby chicks could be reduced by providing them through the first two or three weeks of age with a ration that is heavily fortified with vitamins. It is now the custom to supply chicks with the same starter ration until they are 12 weeks of age or older. In practice it is undesirable to feed an excessive amount of vitamins because rations that carry them are too expensive. On the other hand it is undesirable to feed rations that contain an insufficient quantity of vitamins, because they permit a higher death rate and they also are too expensive. We hope to investigate the advisability, at least when there is any doubt as to the vigor of newly hatched chicks, of supplying them with a repletion ration for the first week or two, and then supplying them with the starter ration only after their vitamin reserves have been restored. According to our working hypothesis the critical vitamin-containing feedstuffs should be incorporated in this repletion ration at the highest levels in order that any vitamin deficiencies may be remedied without delay. Past experience suggests that such a ration should contain from 15 to 25 per cent of soybean meal, 5 or 10 per cent of fish meal, 5 or 10 per cent of dried skim milk and, if available, 5 per cent of dried yeast. There are probably other supplements that would be equally effective. If the supplies of such feedstuffs as dried skim milk are limited the possibility should be explored that the mortality rate could be reduced

by increasing their percentage in the ration during the first three weeks, and then lowering it later on. If the chicks are in an optimum nutritional state at the end of three weeks the reduction would probably not be accompanied by a corresponding increase in the death rate. During this period a rapidly growing chick consumes a relatively small amount of feed, and the higher cost of a more expensive ration would not be a large item. If the mortality rate was markedly reduced the cost per chick would also be reduced.

Pilot Trials. On several occasions poultry growers have come to us with stories of impending disaster. They had flocks of several hundred or thousand chicks (or poults) which with little warning had begun to die at an alarming rate. No infectious disease could be discovered, and our own investigations led us to believe that an unsuspected nutritional deficiency was the cause of the deaths. In no case were we able to identify any specific deficiency, but we were able to demonstrate beyond question that an improved nutritional regime would stop the losses.

It seems that before starting a feeding operation on a scale that could cause financial embarrassment if trouble developed, it would be a helpful practice to run a small scale pilot trial. This test should be run with the same lot of feed that is to be used in the later large scale operation, and should precede it by about 2 weeks. If malnutrition should appear in the pilot flock there would be time to diagnose the difficulty and adopt remedial measures before any serious damage was done.

Significance of Early Deaths. It has happened at times that chicks were distinctly lacking in vitality when they reached the laboratory, and in a very small percentage of the total number of shipments, a large proportion died even though they received a good ration. Usually these unexpected deaths occurred during the first week but occasionally they were distributed over the first two weeks. It is our impression that there are two different causes of these early deaths. One is chilling of the chicks during shipment, or possibily some failure in the operation of the incubators, and after the injury has occurred rescue is impractical, if not impossible. It is our opinion that another important cause of early deaths is a subnormal nutritional state at the beginning of the feeding period. According to our tentative hypothesis these chicks are hatched from eggs which were deficient in certain vitamins, and the chicks are depleted of these vitamins when they leave the shell. They have a poor appetite, consume a subnormal amount of food, and if their rations contain only minimum amounts of the deficient vitamins they die before there is time for the vitamin content of their tissues to rise to a protective level. It has been our custom to disregard all mortalities that occur within the first week after hatching, but it may be that when the adequacy of a ration is under investigation this practice is a mistake. If a group contains

an undue proportion of chicks of low vitality, it may be that many of them could be rescued by a superior diet. To the practical poultryman it is in some degree irrelevant that a ration permits survival when supplied to a college flock that has been carefully selected for vigor, if it fails to permit survival in the chicks he has on hand. It would seem then that when a ration is being tested for adequacy the test should be made with the kind of chicks poultrymen use, rather than with ideal specimens which are least likely to yield useful information. It may be then that mortalities should be counted after a preliminary period that is shorter than one week. This period would end when practically all the chicks had died which could not be rescued from a subnormal nutritional state by attention to the diet. The length of this preliminary period, during which mortalities should not be charged against the feed, has not been established.

BIBLIOGRAPHY

1. Snedecor, G. W., 1940, *Statistical Methods.* Iowa State College Press.

2a. Briggs, Jr., G. M., R. C. Mills, C. A. Elvehjem, and E. B. Hart, 1942, *Nicotinic Acid in Chick Nutrition.* Proc. Soc. Exp. Biol. Med., Vol. 51, pp. 59-61.

2b. Briggs, Jr., G. M., T. D. Luckey, L. J. Teply, C. A. Elvehjem, and E. B. Hart, 1943, *Studies on Nicotinic Acid Deficiency in the Chick.* J. Biol. Chem., Vol. 148, pp. 517-522.

3. Hill, F. W., M. L. Scott, L. C. Norris and G. F. Heuser, 1944, *Deficiency of Unidentified Vitamins in Practical Chick Rations.* Poultry Science, Vol. 23, pp. 253-255.