

ADAPTATION OF ANTIBIOTIC AND VITAMIN B₁₂
FEEDING SUPPLEMENTS TO CHICK DIETS

by

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INTRODUCTION

The rapid development of intensified commercial broiler production has created a tremendous demand for vegetable protein. This demand, in addition to the cost and scarcity of high quality vegetable protein feedstuffs, has prompted research workers to seek ways by which additional sources of vegetable protein can be satisfactorily utilized in chick diets.

Recent investigations, with cottonseed meal as a source of vegetable protein in chick starting rations, indicate that antibiotics and vitamin B₁₂ possess growth stimulating properties when used as supplements to these diets (Sherwood and Couch, 1950). This study and work of Richardson and Blaylock (1950 b) also suggests that further studies regarding antibiotic and vitamin B₁₂ supplementation of cottonseed meal diets might improve the nutritional value of the diets to the extent that a source of vegetable protein in addition to soybean meal could be efficiently utilized.

Using growth as a criterion, two experiments were conducted in an effort to accomplish the following objectives: (1) Compare the nutritional value of cottonseed oil meal and soybean oil meal; (2) Compare the growth response promoted by supplementing both vegetable sources of protein with identical levels of various antibiotic and vitamin B₁₂ feeding supplements; (3) Determine the extent and conditions under

which cottonseed meal can be used in practical starting diets to obtain optimum growth of chicks.

REVIEW OF LITERATURE

Numerous investigations have been conducted studying the growth responses produced by soybean oil meal diets supplemented with antibiotics and vitamin B₁₂. In contrast, the literature reveals comparatively few such growth studies with cottonseed oil meal.

Sherwood and Couch (1950) compared the growth responses produced by soybean and cottonseed oil meal diets when supplemented with a 1 per cent APF¹ concentrate (Lederle Aureomycin fermentation product, designated as "Animal Protein Factor"). This study showed that the cottonseed oil meal diet failed to produce a growth response equivalent to that obtained with a soybean oil meal diet supplemented in an identical manner. Sherwood and Couch concluded that the addition of an APF concentrate, D-L lysine and fish solubles should produce satisfactory growth in chicks when cottonseed oil meal was used as the principal source of vegetable protein. This diet improved the growth rate considerably, but did not equal the growth rate attained by the control diet of soybean meal supplemented similarly with the exception of the D-L-lysine. The

¹The term APF has been officially discontinued as of January, 1951. The terms "vitamin B₁₂" and "antibiotic feeding supplement" replaces the previous term.

soybean oil meal diet, supplemented with APF concentrate was also slightly superior to the more adequately supplemented cottonseed oil meal diet. Heywang and Bird (1950) reported that the amino acid, lysine and probably vitamin B₁₂ was inadequate in cottonseed meal. It was further indicated that APF was not the first limiting factor in the meal, but that it was more deficient in factors found in sardine fish meal. That cottonseed meal is apparently deficient in vitamin B₁₂, was indicated in earlier investigations by Bird and associates (1948) when they concluded that both soybean and cottonseed oil meal were deficient in factors existing in cow manure and fish meal. Using chicks, Ott et al. (1948) investigated the activity of crystalline B₁₂ when fed with soybean meal as the principal source of vegetable protein in a diet. These workers reported that 30 micrograms per kilogram of diet appeared to be the optimum growth requirement for vitamin B₁₂ when used with soybean oil meal alone. Richardson and Blaylock (1950 b) also demonstrated the need for vitamin B₁₂ in an all-vegetable protein diet by comparing a diet containing a commercial cottonseed meal of low gossypol content (.035 per cent) with a diet containing soybean meal. The soybean meal diet supported a normal rate of growth when supplemented with a vitamin B₁₂ concentrate equivalent to 10-20 micrograms of vitamin B₁₂ per kilogram of diet. The cottonseed meal diet produced a comparable growth response when supplemented with

vitamin B₁₂ and 0.2 per cent L-lysine. However, the cottonseed meal diet plus lysine, but without B₁₂, gave a subnormal rate of growth, thus confirming previous workers conclusions that cottonseed meal is deficient in this vitamin. The authors cited, combined the two protein concentrates in a diet supplemented with B₁₂, and obtained growth equivalent to that produced by the diet containing soybean meal alone. Without B₁₂, growth was subnormal. Grau (1950) indicated that under practical conditions, diets containing expeller-type cottonseed meal probably need no amino acid supplement, but recommended that 3 per cent fish meal and a source of vitamin B₁₂ be added to the diet.

The relationship between vitamin B₁₂ and antibiotics has appeared to be only vaguely understood. This is clearly indicated by the tendency of workers in earlier years, 1946-1950, to refer to vitamin B₁₂ and antibiotics interchangeably as the "chick growth" factor, "animal protein" factor, and "cow manure" factor. Patton (1951) reported experiments in which both vitamin B₁₂ and aureomycin were needed for satisfactory growth of chicks fed an all-vegetable protein diet. These experiments revealed no evidence of B₁₂ synthesis resulting from aureomycin in the chick's diet. It was felt that most growth studies with chicks appear to indicate separate requirements for vitamin B₁₂ and an antibiotic when standard diets are used. However, previous work by Cravioto-

Munoz et al. (1951) with rats fed purified diets, demonstrated that aureomycin may replace vitamin B₁₂ completely by producing equal or superior growth responses. In their experiments with swine, Cunha and associates (1951) revealed that certain basal diets supplemented with aureomycin alone were of slightly more value than those supplemented with a 1 per cent aureomycin-B₁₂ product. These workers seemed to believe that the growth promoting effect of various antibiotics varies with the species and the nature of the diet. Jukes et al. (1950) confirmed this hypothesis with their experiment with swine. Groschke and Evens (1950) reported that certain antibiotics lowered the requirements of vitamin B₁₂ in the chick. McGinnis et al. (1950) presented data which suggested that one function of antibiotics may be the "sparing" of vitamin B₁₂. Stokstad and Jukes (1950a) reported that fermentation products of Streptomyces aureofacium promoted growth in chicks depleted of the "animal protein factor" when fed diets which were adequately supplied with vitamin B₁₂. Stokstad and Jukes (1950b) fed crystalline aureomycin HCL with a vegetable protein diet, and reported growth responses in turkey poults with and without vitamin B₁₂. Further work by Stokstad and Jukes (1951) showed that aureomycin had a sparing effect on the B₁₂ requirement in some experiments, but not in others, when chicks were fed diets deficient in vitamin B₁₂ with and without aureomycin supplementation. The aureomycin

promoted growth both in the presence and absence of vitamin B₁₂. Coates and his associates (1951) enlarged upon the theory of "sparing of vitamin B₁₂." Their studies with B complex depleted chicks, revealed that other vitamins were also spared by the addition of antibiotics. Biely and March (1951) reported similar results and concluded that aureomycin lowered the dietary requirements for certain other vitamins of the B complex in the chick; namely, riboflavin, folic acid and niacin.

Some workers have reported no evidences of the sparing effect of antibiotics on vitamin B₁₂. Johnson (1951) stated that feeding antibiotics does not change vitamin requirements. The author cited maintains that the B₁₂ requirements of poultry are much lower than for other B vitamins. Johnson conducted experiments which indicated that the addition of levels as low as 1½ to 2 milligrams per ton of feed produced satisfactory growth in chicks. Davis and Briggs (1951) could not demonstrate an apparent sparing action by various antibiotics on vitamin B₁₂ for either the chick or the poult.

The growth stimulating effect of vitamin B₁₂ has been attributed to increased feed intake, increased efficiency of feed utilization or a combination of these two factors. Rupp et al. (1951) apparently eliminated the factor of increased feed intake by force feeding rats at a constant rate, thus indicating the possibility that vitamin B₁₂ improves the

efficiency of feed utilization, especially protein utilization. Abbott (1951) reported that vitamin B₁₂ may increase the efficiency of utilization of both nitrogen (protein) and phosphorus in the chick. Similiar findings with regard to antibiotics were reported by Thayer et al. (1950) with a study of comparative values of specific vegetable and animal protein concentrates for chicks. Results of Cunha and co-workers (1950), with swine experiments, apparently confirm the work of Thayer and his associates. Davis and Briggs (1951) demonstrated that chicks showed a marked improvement in feed efficiency when the diet was supplemented with an antibiotic. No indication was noted that growth stimulation occurred as a simple manifestation of increased feed consumption.

Workers have attempted to account for the growth stimulatory effect of antibiotics in various experiments. Groschke and Evans (1950) offered the explanation that antibiotics stimulated growth indirectly by altering the intestinal microflora from "undesirable" to "desirable" types of organisms. The unknown factor synthesized by the "desirable" types of microflora appear to be responsible for the increase in growth. McGinnis et al. (1950) suggested that the beneficial effects of the antibiotics are due to the inhibition of the organism *Clostridium (Welchi)*, more recently known as *Clostridium (perfringes)*, which is toxic to the growing chick. Biely and March (1951) hypothesized that antibiotics cause a re-

duction in the number of intestinal microflora which might compete with the host for members of the vitamin B complex. These workers further believe that antibiotics permit the proliferation of microorganisms which synthesize these vitamins. Moore et al. (1946) added antibiotics to a purified diet for chicks and observed that the drug did not cause a sterilization of the intestinal tract, but did produce a marked reduction in the coliform bacteria of the cecal contents.

Coats and associates (1951) concluded that the tendency of antibiotic containing diets, to reduce mortality, suggests the theory that the drug acts by combating a minor infection. However, they postulated that their results with biotin, folic acid and niscin, gave strong evidence in favor of its altering the microbial population of the intestine.

Growth studies have been conducted to determine the stimulatory effect of specific antibiotics in an effort to compare their values as supplements. Newell et al. (1947) reported that, with a practical starting diet, a combination of penicillin mycelium and fish press water resulted in excellent growth of chicks. Berg and coworkers (1950) fed, then deleted, an aureomycin fermentation product from the ration of chicks, and noted that the accelerated growth response observed during the first 4.5 weeks of life ceased.

Birds fed a basal diet with no aureomycin from 1 day old to 4.5 weeks were given the antibiotic, and made slightly greater gains than birds which had received the material from 1 day of age. In comparing aureomycin and streptomycin for chicks and poults, McGinnis and associates, (1950) observed that aureomycin stimulated greater growth responses. Lawrence et al. (1950) added aureomycin fermentation product to a "high energy" fryer ration and noted a significant increase in weight. Johnson (1951) found that penicillin gave a better growth response when fed at a level of 2 to 4 grams per ton than aureomycin at 10 grams per ton. Matterson et al. (1951) compared aureomycin, streptomycin, penicillin, terramycin, and bacitracin as supplements to all-vegetable protein diets. All were added at the level of 9 grams per ton. These workers found that streptomycin was least effective in promoting growth and indicated that the greatest per cent growth response with antibiotics was obtained on poorer quality (all-vegetable protein) rations. The greatest weight was obtained on the better quality rations (all-vegetable protein plus fish meal) supplemented with antibiotics. Further observations by these workers revealed that when the ration contained fish meal, penicillin and bacitracin appeared to react differently from other antibiotics by maintaining for a longer period of time an increased percentage growth response as compared with the

unsupplemented basal.

Research workers conducting growth studies with cottonseed meal in the past have been confronted with the growth depressant effect of the polyphenolic pigment compound, gossypol, found in the pigment glands of the meal. Boatner and associates (1948) observed little retardation in growth of chicks attributed to mixing components of cottonseed pigment glands in diets at low levels. However, the addition of high levels of pure gossypol (in excess of .003 per cent) produced significant suppressions of growth. The results of the investigation by Lillie and Bird (1950) were in complete disagreement with the findings of Boatner et al. (1948) in that these workers noted no significant difference in the toxicity of the two forms of gossypol. These workers administered pure gossypol and gossypol pigment glands daily by capsule at levels which supplied equivalent quantities of both forms of the compound. It was concluded that the toxicity of pigment glands administered in this manner was due entirely to their gossypol content. Heywang et al. (1952) apparently clarified the conflicting opinions by demonstrating that pure gossypol is lost or inactivated when mixed with diets and allowed to stand. Gossypol was mixed with diets at a level of .012 per cent, allowed to stand for 144 hours and then chemically analyzed. Only .003 per cent gossypol was

detectable after the 144 hour period. These workers confirmed the chemical analysis results by conducting growth studies with chicks. The authors cited, concluded that the findings of Boatner and associates (1948) were erroneously influenced by the "loss or inactivation" characteristic of gossypol when mixed in feeds.

The amino acid deficiency of lysine in cottonseed meal has been investigated. Grau (1946) studied amino acid sources in concentrates for chicks, and concluded that the amino acid analysis of cottonseed meal pointed to a deficiency in lysine. Addition of lysine supplements to basal diets increased growth from 4.4 to 5.7 per cent. Heywang and Bird (1950) conducted subsequent experiments which confirmed the findings of Grau. Richardson and Blaylock (1950a) investigated a diet for turkey poults which contained cerelese and 60 per cent cottonseed meal. The diet supported a rapid rate of growth when supplemented with .5 per cent L-lysine. The more recent work of Milligan et al. (1951) provided significant information as to the lysine requirement for chicks fed cottonseed meal.

It appears that a cottonseed meal of high nutritional value can be prepared by an improved screw-press method. According to Altschul (1950) the following are factors which are important in this method of processing: (1) the free

gossypol content should be .003 per cent or lower as determined by the Southern Research Laboratory method;¹

(2) the meal should not be cooked at a temperature exceeding 200°F. The work of Olcott and Fontaine (1941) demonstrated the damaging effect of heat on cottonseed meal. Ingram, Cravens and Elvehjem (1950) measured the biological value of various cottonseed meal samples which were processed by the hydraulic and screw-pressed methods. A comparison of the two samples revealed that with an increase in temperature, the liberation of the amino acids, mainly lysine, by enzymatic hydrolysis was decreased. The chick growth supported by the samples confirmed the results obtained by enzymatic hydrolysis. In the screw-press method, additional heat is applied to the meal at the time of pressing, while very little heat is developed by the hydraulic process. On the other hand, the per cent of free gossypol is considerably lower in the screw-pressed meal than in the hydraulic type.

¹Hopper, T. H. Review of methods of analysis of cottonseed meals. Proceedings of a Research Conference on Processing as Related to Cottonseed Meal Nutrition at Southern Regional Research Laboratory, p. 14. 1950.

MATERIALS AND METHODS

The experiments were conducted in the Poultry Nutrition Laboratory at the College Poultry Farm. Room temperature was controlled by thermostat, maintained between 70 and 75 degrees Fahrenheit, and heated by natural gas.

Each deck of a contact type starting battery was symmetricaly divided through the center with sheet aluminum, thus providing ten compartments with a five deck battery suitable for small experimental lots of chicks.

Temperature, heating-unit height, feeders and waterers were adjusted in keeping with the growth and age of the chicks. The chicks were transferred to growing batteries at the age of five weeks.

Crossbred chicks (New Hampshire X Kansas State College Strain White Plymouth Rock) were used in both experiments. All chicks were wing banded for identification, weighed at one day of age, and every seven days thereafter throughout the duration of the two experiments. Weekly feed consumption records were maintained.

Experiment I

Two-hundred chicks were randomized into ten lots of 20 chicks each.

Two basal diets were prepared. Basal I was that used by the Kansas State College Experiment Station and referred to as

Table 1. Composition of the basal diets for Experiment I.

Ingredients	Basal diet	
	I	II
Ground yellow corn	61.5 lb	58.0 lb
Wheat Bran	4.0	4.0
Alfalfa meal (17% Dehyd.)	1.0	1.0
Soybean oil meal (44% solvent ext.)	30.0	-
Cottonseed oil meal (41% hydraulic)	-	33.0
Calcium carbonate	1.0	2.0
Steamed bone meal	2.0	1.5
Salt (NaCl)	0.5	0.5
Manganese sulfate	25.0 g	5.0 g
"Prot A" ¹	100.0	100.0
"Delsterol" --D ₃ ²	40.0	40.0
Riboflavin	5.0	5.0
Choline chloride (crystalline)	18.0	17.0
Calcium pantothenate	1.0	1.0
Niacin	5.0	5.0
Lysine HCl (anhydrous)	-	90.2
Total	100.0	100.0

Antibiotics and B₁₂ feeding supplements per 100 lb³:

"Aurofac" (1.8 g aureomycin + 1.8 mg B ₁₂ per lb of supplement)	75.6 g
Aureomycin HCl (crystalline)	201.0 mg
"Bacitracin 3-3" (3 g bacitracin + 3 mg B ₁₂ per lb of supplement)	45.4 g
"Bacitracin 5" (5 g bacitracin per lb of supplement)	18.2 g
"Bi-Con 3 + 3" (3 g terramycin + 3 mg B ₁₂ per lb of supplement)	45.4 g
"TM-5 (5 g terramycin per lb of supple- ment)	18.2 g

¹Supplies 2,400 USP units of vitamin A per g of supplement.

²Supplies 2,000 A.O.A.C. units of vitamin D₃ per g of supplement.

³Supplemented at the rate of 10 g of antibiotics and 6 mg of B₁₂ per ton.

the Kansas State High Efficiency all-vegetable protein ration. Antibiotic and vitamin B₁₂ feeding supplements were omitted. Basal II was modified from the Kansas State College diet by substituting 33 per cent of a commercial hydraulic 41 per cent protein cottonseed meal for the soybean meal.

The vitamin, mineral and amino acid content of Basal II was calculated and compared with the National Research Council's Recommended Nutrient Allowance for Poultry (1950). L-lysine monohydrochloride anhydrous¹ was added at a level of 0.2 per cent per 100 pounds of diet. The basal diets are presented in Table 1.

The antibiotics used were: Aurofac (Lederle's) antibiotic feeding supplement containing 1.8 grams of aureomycin fermentation product and 1.8 milligrams of vitamin B₁₂ per pound of supplement; crystalline pure aureomycin HCL (Lederle's); Bacitracin (U.S.I.) antibiotic supplement containing 3 grams of bacitracin fermentation product and 3 milligrams of B₁₂ per pound of supplement; Bacitracin (U.S.I.) antibiotic supplement containing 5 grams of bacitracin fermentation product per pound; Bi-Con- 3 + 3 (Pfizer's) antibiotic supplement containing 3 grams of terramycin fermentation

¹Generously supplied through the courtesy of Merck Co., Rahway, New Jersey.

product and 3 milligrams of B₁₂ per pound, and TM-5 (Pfizer's) antibiotic supplement containing 5 grams of terramycin fermentation product per pound. The amounts added to each basal diet are shown in Table 1.

The basal diets were prepared by weighing the ingredients used in large quantities, on a portable platform scales, and the ingredients used in smaller quantities on a metric trip scale or analytical balance. The minerals were pre-mixed using ground corn and wheat bran in a small feed mixer at the Kansas State College Poultry Farm for 15 minutes. The vitamins were pre-mixed using ground corn in a small closed container, and added to the basic feed ingredients in the large horizontal-type batch mixer along with the mineral mix for 30 minutes. The two basal diets were mixed separately and five equal portions of each were weighed out. The various antibiotic feeding supplements were then added to eight diets, each being mixed in the small mixer for 15 minutes. The two remaining portions of each basal were fed as such. The crystalline aureomycin HCL was dissolved in a water solution. A 10 cc pipette was used to distribute the liquid as a fine spray while the diet was being mixed. The diets were stored in tightly covered metal containers and were placed in the Poultry Nutrition Laboratory. The remainder of the diets was stored in the feed room at room temperature and transferred later to the laboratory as needed. The diets were prepared

three times during the experimental feeding period. The experiment was terminated when the chicks were eight weeks of age.

Experiment II

Sexed chicks were used in this experiment, 11 males and 9 females in each of 10 lots.

The cottonseed meal included in Basals III, IV, V and VI, was an improved screw-pressed type, experimentally processed under the following specific conditions: maximum temperature in cooker 200° F., moisture content of meats leaving cooker 4 per cent, and free gossypol content .035 per cent.¹ The hydraulic type meal was processed as follows: maximum temperature in cooker 230° F., moisture content of meats leaving cooker 8.4 per cent, and gossypol content .055 per cent.² The composition of each basal is shown in Table 2.

The L-lysine (monohydrochloride, anhydrous) was supplemented at a level of 4.25 grams per pound of cottonseed meal.³

¹Generously supplied by the Southern Regional Research Laboratory, New Orleans, La.

²Gossypol analysis conducted through the courtesy of the Protein and Carbohydrate Division of the Southern Regional Research Laboratory.

³Personal Communication: Ltr. from Dr. F. H. Thurber, chemist, Southern Regional Research Laboratory.

In view of findings in the previous experiment, the combination antibiotic supplement, Aurofac-Bacitracin was used and added to the basals at the same level as in Experiment I.

All diets were mixed in the small mixer, as previously described. Basal II was first mixed and weighed out into two equal portions. The antibiotic and B₁₂ supplements were added to the diet for Lot 5 and mixed for 15 minutes. The portion for Lot 2 was fed as such. The same procedure was followed in preparing the diets containing the screw-pressed meal fed Lots 3 and 6. The previously described procedure was also applied in the preparation of the Kansas State College Basal I diet for Lots 1 and 4. The diets fed Lots 7 and 8 as Basal IV were prepared and mixed as one, the lysine supplement was omitted. The equal portions were weighed out with lysine added to the portion fed Lot 8. The diets for Lots 9 and 10, fed as Basal V and VI, respectively, were prepared and mixed separately.

Storage conditions were the same as in Experiment I. The diets were prepared three times during the experimental period. The experiment was terminated when the chicks were eight weeks of age.

Table 2. Composition of the basal diets for Experiment II.

Ingredients	Basal diet					
	I	II	III	IV	V	VI
Ground yellow corn	61.5 lb	58.0 lb	58.0 lb	59.5 lb	66.0 lb	63.0 lb
Wheat bran	4.0	4.0	4.0	4.0	4.0	4.0
Alfalfa meal (17% Dehyd.)	1.0	1.0	1.0	1.0	1.0	1.0
Soybean oil meal (44% solvent ext.)	30.0	-	-	12.0	-	10.0
Cottonseed oil meal (41% screw-pressed)	-	-	33.0	20.0	15.0	15.0
Cottonseed oil meal (41% hydraulic)	-	33.0	-	-	-	-
Meat scraps (50% protein)	-	-	-	-	6.0	2.0
Fishmeal (60% protein)	-	-	-	-	6.5	3.0
Calcium carbonate	1.0	2.0	2.0	1.5	1.0	1.5
Steamed bone meal	2.0	1.5	1.5	1.5	-	-
Salt (NaCl)	0.5	0.5	0.5	0.5	0.5	0.5
Manganese sulfate	25.0 g	5.0 g	5.0 g	5.0 g	5.0 g	5.0 g
"Prot A"1	40.0	40.0	40.0	40.0	40.0	40.0
"Delsterol" D ₃ ²	25.0	25.0	25.0	25.0	25.0	25.0
Riboflavin	5.0	10.0	10.0	10.0	10.0	10.0
Choline chloride (crystalline)	18.0	17.0	17.0	17.0	21.0	18.0
Calcium pantothenate	1.0	1.0	1.0	1.0	1.0	1.0
Niacin	5.0	5.0	5.0	5.0	5.0	5.0
Lysine HCl (anhydrous)	-	140.2	140.2	-	-	-
Total (lb)	100.0	100.0	100.0	100.0	100.0	100.0

Antibiotic and B₁₂ feeding supplement per 100 lb³:

"Auroreac" (1.8 g aureomycin + 1.8 mg B₁₂ per lb of supplement)

Aureomycin HCl (crystalline)

Bacitracin (3 g bacitracin + 3 mg B₁₂ per lb of supplement)

Becitracin (5 g bacitracin per lb of supplement)

¹Supplies 5,000 USP units of vitamin A per g of supplement.

²Supplies 2,000 A.O.A.C. units of vitamin D₃ per g of supplement.

³Supplemented at the rate of 10 g of antibiotics and 6 mg of B₁₂ per ton.

RESULTS

Experiment I

Growth at Eight Weeks The average weight per lot, mortality, and diets fed are presented in Table 3. Growth curves are shown in Figs. 1, 2, 3 and 4. Analysis of variance and "t" test comparisons are found in Tables 4, 5 and 6, respectively.

Growth of chicks receiving the cottonseed oil meal diets was inferior at all ages to those receiving the soybean oil meal diets. An analysis of variance showed a significant difference in the growth produced by the two vegetable proteins used in the basal diets. The chicks fed the cottonseed oil meal basals were more uniform than those receiving the soybean oil meal basals as shown by the standard error of the mean for lot weights.

The growth of all lots receiving antibiotic and B₁₂ feeding supplements was superior to that of lots receiving nonsupplemented basals. An analysis of variance test as shown in Table 4 revealed a highly significant difference in the growth of the birds receiving supplemented and nonsupplemented basal diets. A consistent growth response was noted as a result of the antibiotic and B₁₂ supplementation. As is revealed in Table 3, lots fed the soybean meal basal, supplemented with the Aurofac-Bacitracin combination produced the heaviest lot mean weight of 1065.78 ± 38.03 grams at eight weeks. The mean weight of the heaviest lot receiving the cottonseed meal basal was 755.55 ±

Table 3. Experiment I, diets, average weight and mortality for chicks at eight weeks.

Lot	Diet	Mean Weight in grams	Mortality %
1	Basal I	771.30 ± 40.64	0
2	Basal II	536.27 ± 20.98	5
3	Basal I + Aurolac	1013.65 ± 37.39	0
4	Basal II + Aurolac	629.05 ± 19.65	0
5	Basal I + Bacitracin	989.45 ± 31.24	0
6	Basal II + Bacitracin	658.30 ± 20.12	0
7	Basal I + Terramycin	999.78 ± 32.42	0
8	Basal II + Terramycin	718.65 ± 22.98	0
9	Basal I + Aurolac-Bacitracin	1065.78 ± 38.03	5
10	Basal II + Aurolac-Bacitracin	755.55 ± 15.07	0

Table 4. Experiment I, analysis of variance of growth at eight weeks.

Source of variation:	Degrees of freedom	Sum of squares	Mean square
Antibiotics	4	1,515,188	378,797 **
Protein	1	4,745,224	4,745,227 **
Sex	1	713,531	713,531 **
Interactions	13	456,805	35,139 **
Individuals	178	2,120,467	11,913
Total	197	9,551,215	

15.07. This lot also received the combination antibiotic and B₁₂ supplement.

Growth of the lots receiving the cottonseed meal basal was more uniform, as shown by the relatively small standard error of the mean. This difference in uniformity of growth produced by each basal was consistent in all lots of birds.

Under the conditions of this experiment, few significant differences were noted as a result of comparing growth promoted by different antibiotic and B₁₂ supplements. A study of Table 5 reveals that, on a lot mean basis, no significant differences were observed when the lots fed Basal I supplemented with antibiotics and B₁₂ were compared. On the other hand, a highly significant difference was noted when the mean weight of birds in Lot 4, Basal II (Aurofac), and Lot 10, Basal II (Aurofac-Bacitracin) were compared. The mean weight of birds in Lot 10 was greater by a margin of 126.50 ± 24.76 grams, a highly significant increase. The birds in Lot 10 also exceeded those in Lot 6 receiving Basal II (Bacitracin). This lot had a mean weight of 755.55 ± 15.00 grams as compared with 658.30 ± 20.12 grams for Lot 6. The 97.25 ± 25.13 gram margin was highly significant. There were no further significant differences observed when other lots were compared. When the data were analyzed according to the mean weights of each sex within the lots, and each lot compared, a significant increase was noted

Table 5. Experiment I, a comparison of lot mean weights of chicks at eight weeks.^a

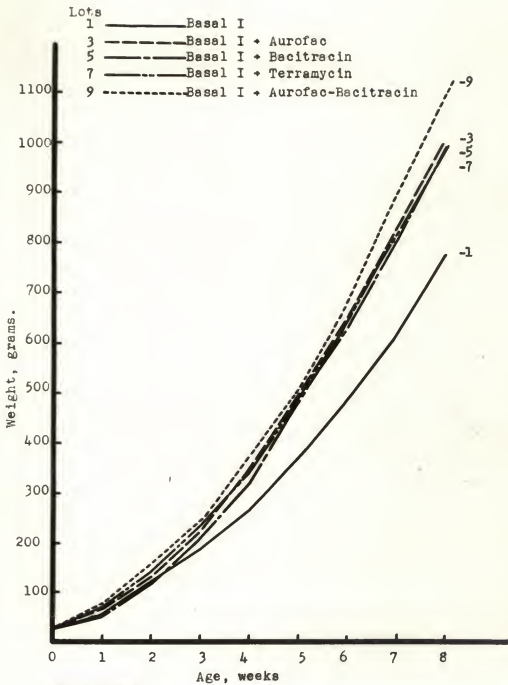
Lot:	Basal:	Supplement	Mean Weight in grams:	Difference:	Value of t.
9	I	Aurofac-Bacitracin	1065.78 ± 38.03		
3	I	Aurofac	1013.65 ± 37.39	52.13	53.33
10	II	Aurofac-Bacitracin	755.55 ± 15.07		
4	II	Aurofac	629.05 ± 19.65	126.50	24.70 **
9	I	Aurofac-Bacitracin	1065.78 ± 38.03		
5	I	Bacitracin	989.45 ± 31.24	76.33	49.22
10	II	Aurofac-Bacitracin	755.55 ± 15.07		
6	II	Bacitracin	658.30 ± 20.12	97.25	25.13 **
5	I	Terramycin	999.78 ± 32.42		
7	I	Bacitracin	989.45 ± 31.24	10.33	45.02
8	II	Terramycin	718.65 ± 22.98		
6	II	Bacitracin	658.30 ± 20.12	60.35	30.34

^a Lots fed the eight supplemented basals.

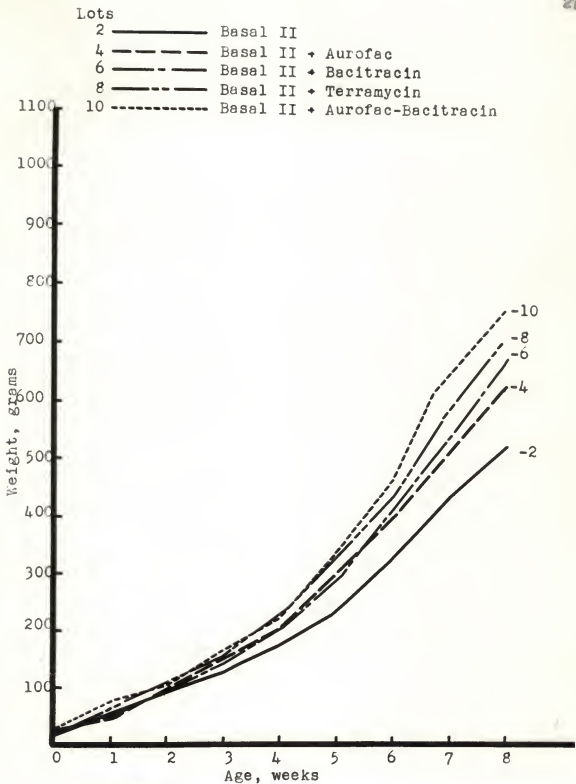
Table 6. Experiment I, a comparison of mean weight of chicks by sex at eight weeks.^a

Lot	: Basel	: Supplement	: Mean Weight	: Differ- ence	: Value of t.
Males					
3	I	Aurofac	1192.00 ± 22.32		
5	I	Bacitracin	1066.75 † 52.93	125.25	62.68*
6	II	Bacitracin	684.28 † 39.13		
4	II	Aurofac	680.66 † 30.65	3.62	49.71
9	I	Aurofac-Bacitracin	1173.70 ± 41.95		
7	I	Terramycin	1062.14 † 48.07	111.56	63.80
8	II	Terramycin	754.00 † 18.14		
10	II	Aurofac-Bacitracin	771.77 ± 56.56	12.23	58.55
3	I	Aurofac	1192.00 ± 22.32		
7	I	Terramycin	1062.14 ± 48.07	129.86	53.00*
Females					
9	I	Aurofac-Bacitracin	945.88 † 35.41		
3	I	Aurofac	894.75 † 77.55	51.13	85.25
10	II	Aurofac-Bacitracin	742.24 ± 37.42		
4	II	Aurofac	606.92 † 26.91	135.35	46.08 **
9	I	Aurofac-Bacitracin	945.88 ± 35.41		
5	I	Bacitracin	937.91 ± 27.98	7.97	45.13
10	II	Aurofac-Bacitracin	742.27 ± 37.42		
6	II	Bacitracin	644.30 † 23.30	97.97	44.07*
8	II	Terramycin	683.46 ± 29.44		
4	II	Aurofac	606.92 ± 26.91	76.54	39.96

^aLots fed the eight supplemented basals.

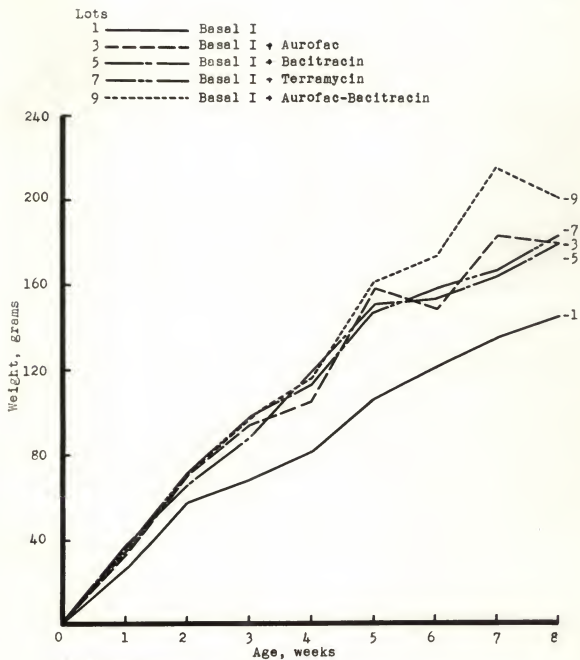


Experiment I
 Fig. 1. Growth of chicks, 5 lots.



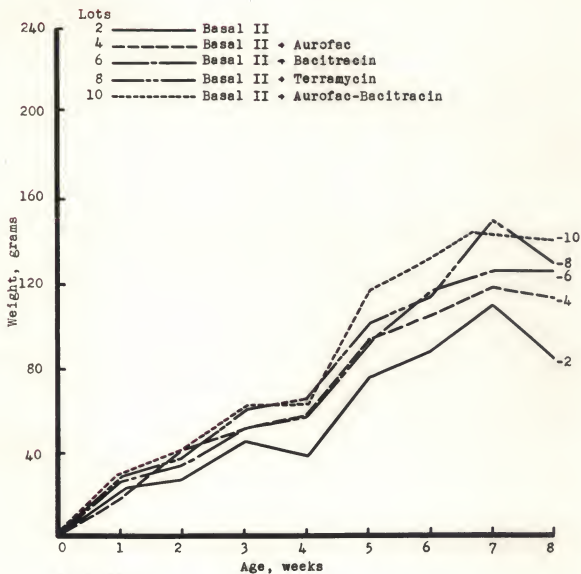
Experiment I

Fig. 2. Growth of chicks, 5 lots.



Experiment I

Fig. 3. Average gain of chicks, 5 lots.



Experiment I

Fig. 4. Average gain of chicks, 5 lots.

for the male birds in Lot 3 fed Basal I, (Aurofac) in relation to male birds in Lot 7 receiving Basal I, (Terramycin). Table 6 reveals a margin of 129.86 ± 53.00 grams. A further study of Table 6 shows that other comparisons indicate differences that approach significance. It was observed that a highly significant increase prevailed within the lots of birds fed Basal II when analyzed according to sex. Again, the combination, Aurofac-Bacitracin supplemented basal (Lot 10), produced heavier birds when females were compared with those in Lot 4 receiving Basal II (Aurofac). The female mean weight of Lot 10 was 742.27 ± 37.42 grams as compared with 606.92 ± 26.91 , (female mean weight of Lot 4), the difference of 135.35 ± 46.07 grams being highly significant. The mean weight of the females in Lot 10 was also significantly greater than that of the birds in Lot 6, receiving Basal II (Bacitracin). Further study of Table 6 shows that other differences approached significance.

Weekly gain was greater in lots fed basals supplemented with antibiotics and B₁₂ than those fed nonsupplemented basals, as shown by the graphs in Fig. 2 and 3. It was noted that gains of chicks in lots receiving the Aurofac-Bacitracin supplemented basals was markedly reduced between the seventh and eighth week.

Feed Efficiency. Feed efficiencies were calculated on a chick-day basis as grams of feed required per gram of gain. Weekly efficiencies for each lot and average Fig. are presented in Table 7.

Table 7. Experiment I, Feed efficiency (gm. feed per gm. gain)

Lot no.	Weeks								Average
	1	2	3	4	5	6	7	8	
1	2.44	2.07	2.74	2.91	2.88	2.75	2.47	3.25	2.67
2	3.23	3.70	3.12	4.32	3.12	3.45	2.94	3.76	3.46
3	2.15	1.95	2.08	2.07	2.28	2.51	2.95	2.99	2.37
4	3.46	2.55	3.36	3.38	2.81	2.95	2.91	3.42	3.10
5	1.36	1.99	2.64	2.19	2.41	2.62	2.88	2.86	2.37
6	1.82	3.00	3.11	3.38	2.81	2.72	3.30	3.47	2.95
7	2.34	1.87	1.68	2.39	2.43	2.79	2.88	2.79	2.42
8	2.75	2.99	2.65	4.09	2.51	2.53	2.62	3.01	2.89
9	1.36	1.96	2.01	2.39	2.49	2.62	2.68	6.65	2.77
10	3.03	4.24	2.78	3.67	2.68	2.97	3.00	3.42	3.22

Chicks receiving the soybean meal basal utilized feed more efficiently than those receiving cottonseed meal diets. The chicks receiving basals supplemented with antibiotics and B₁₂ converted feed more efficiently than those receiving the basals alone, with two exceptions, the chicks receiving the Aurofac-Bacitracin combination supplement in both basals were slightly less efficient in their conversion. These lots on both basals attained the greatest average weight of all diets used in the experiment. Chicks receiving Basal I with antibiotic and B₁₂ supplements added, converted feed more efficiently than did those receiving Basal II supplemented with antibiotics and B₁₂.

Mortality. Mortality was negligible during the experiment as indicated in Table 3. Only 2 chicks were lost during the experiment. One died in Lot 2 at 11 days of age. A necropsy revealed no abnormalities or symptoms of a pathological condition. One chick in Lot 7 was accidentally smothered to death.

Other Observations. The overall appearance of the birds receiving the soybean meal basal was better than those fed the cottonseed basal as shown by a study of Plates I, II, and III. The skeletal structure of the birds fed Basal I appeared to be more fully developed than those fed Basal II. The feathers of the birds on the soybean meal diets developed earlier than those fed the cottonseed meal diets. Between the sixth and

eight week the chicks fed the nonsupplemented cottonseed meal basal seemed to "tire" of the diet, weekly consumption remaining constant in contrast to increasing consumption of feed by the chicks receiving both the nonsupplemented Basal I and Basal I and II supplemented with antibiotics and B₁₂.

Several chicks were observed which had symptoms characteristic of riboflavin deficiency. The "curled toe" condition appeared in the eighth week of the experiment. The condition was found only in lots receiving Basal II, irrespective of supplementation.

EXPLANATION OF PLATE I

Males from various lots in Experiment I. Chicks pictured are those nearest the lot mean weight at eight weeks.

- Fig. 1. This male received Basal I, unsupplemented, and weighed 876 grams.
- Fig. 2. This male received Basal II, unsupplemented, and weighed 539 grams. Note that body size of this bird is smaller than that of the bird in Fig. 1.
- Fig. 3. This male received Basal I plus Aurofac, and weighed 1194 grams.
- Fig. 4. This male received Basal II plus Aurofac; weight, 703 grams. Note the difference in appearance and body size as compared with the bird in Fig. 3.

PLATE I



Fig. 1



Fig. 2



Fig. 3



Fig. 4

EXPLANATION OF PLATE II

Males from various lots in Experiment I. Chicks pictured are those nearest the lot mean weight at eight weeks.

- Fig. 1. This male received Basal I, plus Bacitracin; weight 1064 grams.
- Fig. 2. This male received Basal II, plus Bacitracin; weight 696 grams. Note the difference in size of legs and body as compared with the bird in Fig. 1.
- Fig. 3. This male received Basal I, plus Terramycin; weight, 1067 grams.
- Fig. 4. This male received Basal II, plus Terramycin; weight, 802 grams. Note the size of legs and body as compared with the bird in Fig. 3.

PLATE II



Fig. 1



Fig. 2



Fig. 3



Fig. 4

EXPLANATION OF PLATE III

Males from various lots in Experiment I. Chicks pictured are those nearest the lot mean weight at eight weeks.

- Fig. 1. This male received Basal I, plus Aurofac-Bacitracin and weighed 1174 grams.
- Fig. 2. This male received Basal II, plus Aurofac-Bacitracin; weight 767 grams. The same difference in size of legs and body can be noted as in Plates I and II.
- Fig. 3. This bird received Basal I, plus Terramycin and was the heaviest female of all lots in Experiment I, weight, 1117 grams.
- Fig. 4. This bird received Basal I, plus Aurofac-Bacitracin; heaviest male of all lots; weight, 1430 grams.

PLATE III



Fig. 1



Fig. 2



Fig. 3



Fig. 4

Experiment II

Growth at Eight Weeks. The average weight of each lot, mortality and diets fed are presented in Table 8. Growth curves are presented in Fig. 5, 6, 7, and 8. Analysis of variance and standard error of the difference comparisons are shown in Tables 9, 10 and 11.

Growth of chicks receiving unsupplemented cottonseed meal Basals II and III was inferior at all ages observed to those receiving Basal I, the unsupplemented soybean meal diet. The low standard error of the mean weights of chicks in lots receiving Basals II and III indicated uniformity of growth, a characteristic which was lacking in the lot of chicks fed unsupplemented Basal I.

Although the chicks used were of the same strain as in Experiment I, a comparison of the results in the two experiments was not justified due to the onset of Newcastle disease at 5½ weeks of age. However, the general results of both investigations were in agreement as shown by analysis of variance (Tables 9 and 10), since the protein sources and antibiotics again produced significant variations in growth.

Under the conditions of this experiment, it was found that Basal III, supplemented with antibiotics, resulted in growth superior to Basal II supplemented in the same manner. The lot mean weight of the birds fed the former diet was

Table 8. Experiment II, diets, average weight, and mortality for chicks at eight weeks.

Lot:	:Mean weight :		% : Mortality
	: in grams		
1 Basal I	636.77	± 48.29	35
2 Basal II	488.36	± 20.66	30
3 Basal III	507.62	± 37.44	35
4 Basal I + Aurolac-Bacitracin	906.00	± 22.61	20
5 Basal II + Aurolac-Bacitracin	623.18	± 10.91	15
6 Basal III + Aurolac-Bacitracin	732.29	± 28.05	30
7 Basal IV + 12% soybean meal + Aurolac-Bacitracin	710.44	± 28.23	20
8 Basal IV + 12% soybean meal + Aurolac-Bacitracin + .187% lysine	785.25	± 39.08	20
9 Basal V + 6% Meat scraps + 6.5% fish meal + Aurolac-Bacitracin	877.71	↑ 10.00	30
10 Basal VI + 2% Meat scraps + 3% fish meal + 10% soybean meal + Aurolac-Bacitracin	909.93	± 48.01	25

Table 9. Experiment II, analysis of variance of growth for chicks in 6 lots at eight weeks.

Source of variation:	Degrees of freedom	Sum of squares	Mean squares
Antibiotics	1	1,710,578	1,710,528 **
Protein	2	784,593	392,296 **
Sex	1	279,715	279,715 **
Interaction	7	3,932	561
Individuals	75	245,175	3,269
Total	86	3,033,993	

Table 10. Experiment II, analysis of variance of growth for chicks in 4 lots at eight weeks.

Source of variation:	Degrees of freedom	Sum of squares	Mean squares
Protein	1	326.666	326.666 **
Sex	1	515,075	515,075 **
Interaction	1	41,942	41,942
Individuals	57	719,744	12,627
Total	60	1,603,427	

732.29 \pm 28.05 grams as compared with 623.18 \pm 10.91 for the lot mean weight of the latter as shown in Table 11. The difference of 109.11 \pm 29.82 grams was highly significant. No significant difference was noted in the lot mean weights of Basals II and III with no antibiotics added. On the other hand, an analysis of the data, (Table 12), comparing females, indicated a significant difference of 75.57 \pm 29.56 grams in the mean weights of female birds fed Basal III compared with those fed Basal II.

The addition of .187 per cent L-lysine and 12 per cent soybean meal to the diet, (Basal IV, Lot 8) did not promote a significant increase in the mean weight of birds in this lot as compared with chicks receiving Basal IV, Lot 7, supplemented with soybean meal, lysine omitted. A study of the data presented in Table 11 reveals a similar result when the mean weight of chicks receiving Basal IV in Lot 8 was compared with that of the birds fed Basal III in Lot 6. The birds in Lot 6 also showed a nonsignificant increase in mean weight when compared with the birds receiving Basal IV, Lot 7.

Considering the conditions of this experiment, the addition of two sources of animal protein (meat and bone scraps and fish meal) to the screw-pressed cottonseed meal

Table 11. Experiment II. A comparison of certain mean lot weights of chicks at eight weeks.

Lot:	Basal:	Supplement	Mean weight	Difference	Value of t.
3	III	None	507.62 ± 37.44		
2	II	None	488.36 ± 20.66	19.26	42.97
6	III	A-B*	732.29 ± 28.05		
5	II	A-B	623.18 ± 10.91	109.11	29.82**
8	II	A-B + SBOM** lysine	785.25 ± 39.08		
7	IV	A-B + SBOM	710.44 ± 28.23	74.81	48.20
10	VI	A-B + SBOM + AP***	909.93 ± 48.01		
4	I	A-B	906.00 ± 22.61	3.93	53.07
10	VI	A-B + SBOM + AP	909.93 ± 48.01		
8	IV	A-B + SBOM + lysine	785.25 ± 39.08	124.68	61.90*
9	V	A-B + AP	877.71 ± 10.00		
7	IV	A-B + SBOM	710.44 ± 28.23	167.27	29.90**
10	VI	A-B + SBOM + AP	909.93 ± 48.01		
9	V	A-B + AP	877.71 ± 10.00	32.22	61.90
9	V	A-B + AP	877.71 ± 10.00		
6	III	A-B	732.29 ± 28.05	145.20	29.76**
8	IV	A-B + SBOM + lysine	785.25 ± 39.08		
6	III	A-B	732.29 ± 28.05	52.96	48.10
6	III	A-B	732.29 ± 28.05		
7	IV	A-B + SBOM	710.44 ± 28.23	11.85	39.78

*A-B Aurofac- Bacitracin

** SBOM Soybean Oil meal

*** AP Animal Protein -- 50% protein meat and bone scraps, 60% protein fish meal.

basal, supplemented with antibiotics and B₁₂, promoted growth on a lot mean basis, significantly superior to all diets used with one exception. This diet was Basal I, supplemented with antibiotics and B₁₂. The lot mean weight of birds given Basal VI, containing 2 per cent meat and bone scraps, 3 per cent fish meal and 15 per cent soybean meal (Lot 10) was 909.93 ± 48.01 grams, (Table 11), as compared with 906.00 ± 22.61 grams for the lot mean weight of birds receiving Basal I with antibiotics (Lot 4). The difference of 3.93 grams was nonsignificant. When the mean weights of males and females in each lot were compared, the males of Lot 10 were heavier than those in Lot 4 by a margin of 59.42 ± 24.90 grams, a significant difference. On the other hand, the mean weight of the females in Lot 4 was greater by 50.02 ± 42.72 grams than the females in Lot 10. The increase was nonsignificant in view of the large standard error of the difference. Further evidence to support the evaluations of this diet is presented in Table 12.

A study of the data included in Fig. 7 and 8 reveals that the average weekly gains of birds fed nonsupplemented basals were consistently less than those of chicks fed basals supplemented with antibiotics, or antibiotics and animal protein. The chicks receiving the all vegetable protein diets, Basal IV with soy-

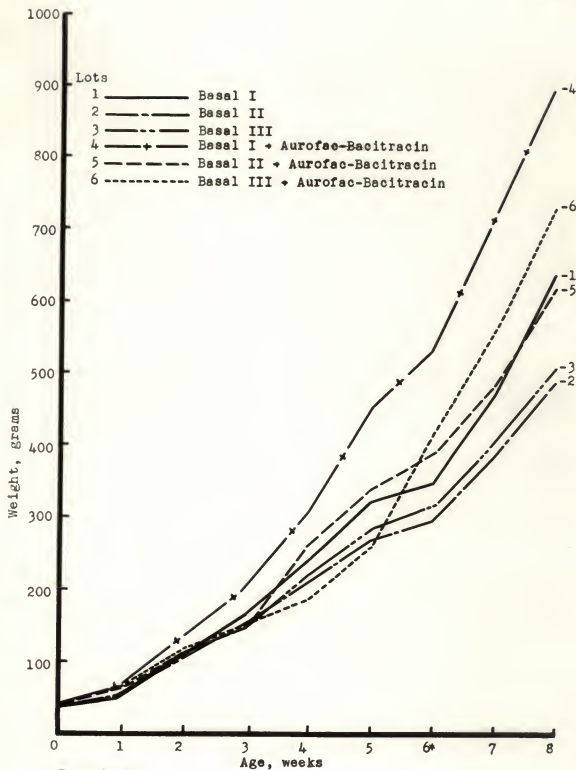
Table 12. Experiment II. A comparison of certain mean weights of chicks by sex at eight weeks.

Lot	Basal	Supplement	Mean Weight	Difference	Value of t.
Males					
2	II	None	521.63 ± 27.20		
3	III	None	493.67 ± 14.93	27.96	14.93
10	VI	A-B* + SBOM** + AP***	1007.33 ± 17.75		
9	V	A-B + AP	934.32 ± 26.76	73.60	32.11*
10	VI	A-B + SBOM + AP	1007.33 ± 17.35		
4	I	A-B	947.91 ± 17.86	59.42	24.90*
8	IV	A-B + SBOM + lysine	868.14 ± 48.08		
7	IV	A-B + SBOM	777.38 ± 23.35	90.76	54.44
Females					
3	III	None	519.57 ± 18.87		
2	II	None	444.00 ± 22.76	75.57	29.56*
9	V	A-B + AP	775.80 ± 40.09		
10	VI	A-B + SBOM + AP	763.83 ± 42.39	11.97	58.35
4	I	A-B	813.80 ± 11.31		
10	VI	A-B + SBOM + AP	763.83 ± 42.39	50.02	42.72
8	IV	A-B + SBOM + lysine	720.78 ± 50.52		
7	IV	A-B + SBOM	643.50 ± 39.89	77.28	64.37

* A-B Aurofac-Bacitracin

** SBOM Soybean oil meal

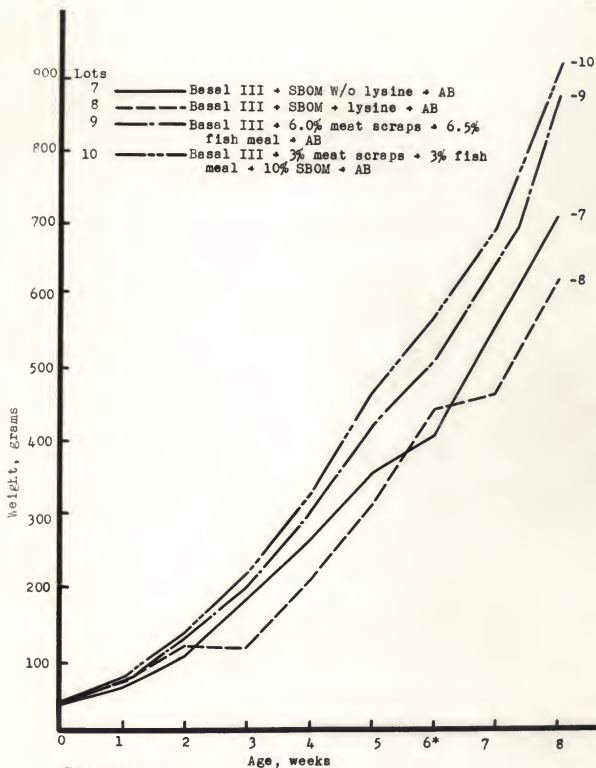
*** AP Animal Protein -- 50% protein meat and bone scraps, 60% protein fish meal.



Experiment II

Fig. 5. Growth of chicks, 6 lots.

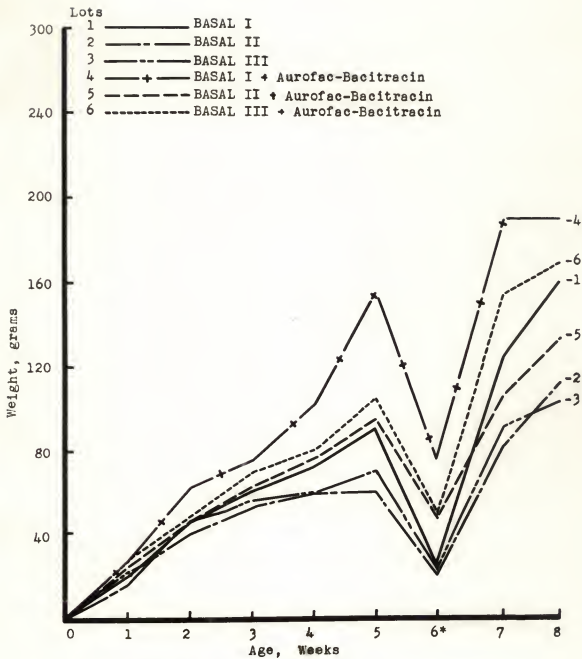
*Onset of Newcastle between 5th and 6th week.



Experiment II

Fig. 6. Growth of chicks, 4 lots.

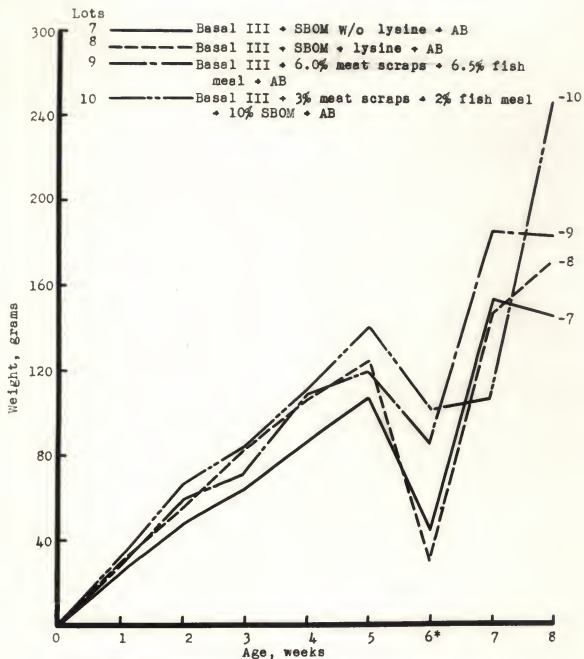
*Onset of Newcastle between 5th and 6th week.



Experiment II

Fig. 7. Average gain of chicks, 6 lots.

*Onset of Newcastle between 5th and 6th week.



Experiment II

Fig. 8. Average gain of chicks, 4 lots.

*Onset of Newcastle between 5th and 6th week.

bean meal plus antibiotics, and Basal IV with soybean meal, antibiotics and lysine, also made greater gains than did the unsupplemented basals. A further study of Figs. 7 and 8 shows that birds in Lots 4, 9, 7, 6, 8 and 5, in the order named, made the most rapid gains after the onset of Newcastle between the sixth and seventh week. Birds in Lots 5 and 10 gained the same during that period. However, birds in Lot 10 gained an average of 249.9 grams between the seventh and eighth week, this gain being the greatest of all lots for the eighth week. The sixth week averages reflect a departure from the normal trend of gain noted during the first and fifth week period. The birds in Lot 10 again maintained the highest average gain of 102 grams for the sixth week. The gains made by the birds receiving the nonsupplemented basals were less than any of the birds receiving other diets during the same period.

Feed Efficiency. Chicks receiving the nonsupplemented basals gave poorer feed efficiency conversions than those receiving the supplemented basals. As is reflected by Table 13 the most efficient of all lots of birds were those in Lot 9 receiving Basal V. A further study of the table reveals that the birds in Lot 10 receiving Basal VI consistently required less feed per gram of gain from the first through the fifth week of the experiment than did the birds in any of the other lots. The birds in Lot 9 and 10 as compared with other lots

Table 13. Experiment II, feed efficiency (gm. feed per gm. gain)

Lot	Weeks								Average
	1	2	3	4	5	6*	7	8	
1	3.82	2.17	2.38	2.61	2.85	9.79	2.19	3.23	3.62
2	2.79	3.47	2.71	2.67	3.41	9.18	2.79	2.94	3.78
3	2.70	2.96	2.55	3.44	3.24	9.18	3.97	3.84	3.99
4	2.51	2.16	2.30	2.74	2.33	4.96	2.26	2.94	2.78
5	2.65	2.52	2.78	3.18	2.09	6.62	3.14	3.88	3.48
6	1.31	3.73	2.60	3.29	2.77	6.18	2.65	3.14	3.21
7	1.36	2.27	2.41	2.19	2.57	6.07	2.36	3.17	2.80
8	2.27	2.39	2.30	2.66	2.56	9.48	2.43	2.93	3.38
9	2.27	2.27	2.30	2.50	2.65	3.90	2.08	3.34	2.66
10	2.05	2.00	2.24	2.69	2.43	4.07	4.55	2.00	2.75

* Onset of Newcastle between the fifth and sixth week.

needed less feed per gram of gain in the sixth and eighth week periods. The birds in Lot 4 receiving Basal I supplemented with antibiotics and B₁₂ also maintained a favorable feed efficiency in the previously mentioned periods. The effect of the incidence of Newcastle disease on the efficiency of feed utilization is clearly evident, as shown by the unfavorable figures for the sixth week period.

Mortality. During the period from the first through the fifth week of the experiment, only one bird died. This chick was in Lot 2 and was lost on the 14th day of the experiment. A necropsy revealed a hemorrhagic condition of the body cavity and bruises on the outside of the body which indicated the chick had been crushed during the night. No pathological conditions were noted.

At 5½ weeks of age, the birds in all lots began sneezing. On the 40th day of the experimental period, 36 hours after the onset of sneezing, two birds died. The birds in all lots were listless, feathers were drooped, and several were observed with heads down. Nine birds died on the 41st day, 12 on the 42nd, 7 on the 43rd, 5 on the 44th, 4 on the 45th, 2 on the 46th, 1 on the 47th, 4 on the 51st, 2 on the 53rd and 1 on the 54th day. The total mortality loss from Newcastle was 51, or 25.6 per cent. Necropsies of several birds revealed congested air sacs and pus-like plugs in the tracheae. Results of the

hemagglutination-inhibition blood test for Newcastle was positive, indicating the presence of antibodies in the blood serum of the chicks.

The feed consumption of the birds in all lots was markedly reduced, and several birds lost weight. Recovery was rapid, but several of the bird's necks remained crooked. At the end of the experimental period, the general health and nutrition of the birds appeared satisfactory.

Other Observations. The general appearance of the birds fed the supplemented basals was excellent. As shown in Plates IV, V, and VI, on a comparable basis, the birds receiving the nonsupplemented basals were not fully developed. The skeletal structure of the chicks fed diets supplemented with animal protein appeared to be more fully developed than those in other lots. At no time during the period before the occurrence of Newcastle were any abnormalities or symptoms of nutritional deficiencies observed. The lots of birds attaining the heaviest weights consumed the greatest amount of feed. Feed consumption appeared to be normal at the end of the period. The birds in Lot 10 consumed more feed than any birds in other lots, a total of 499.4 grams of feed per bird.

EXPLANATION OF PLATE IV

Males from various lots in Experiment II. Chicks pictured are those nearest the lot mean weight at eight weeks.

- Fig. 1. This male received Basal I, unsupplemented; weight, 636 grams.
- Fig. 2. This male received Basal III, (screw-pressed cottonseed meal) unsupplemented; weight, 573 grams.
- Fig. 3. This male received Basal II, (hydraulic cottonseed meal) plus Aurofac-Bacitracin; weight, 623 grams.
- Fig. 4. This male received Basal II, (Hydraulic cottonseed meal) unsupplemented; weight, 488 grams. Note the difference in the leg and body size of birds in Figures 1, and 2, as compared with that of the bird in this Figure.

PLATE IV



Fig. 1



Fig. 2



Fig. 3



Fig. 4

EXPLANATION OF PLATE V

Males from various lots in Experiment II. Chicks pictured are those nearest the lot mean weight at eight weeks.

- Fig. 1. This male received Basal V, (screw-pressed cottonseed meal) + 6% meat scraps, + 6.5% fish meal and Aurofac-Bacitracin; weight 877 grams.
- Fig. 2. This male received Basal III, (screw-pressed cottonseed meal) + Aurofac-Bacitracin; weight 732 grams. Note the difference in body size as compared with birds in Figures 1 of this plate, and Figures 2, 3, and 4 of Plate IV.
- Fig. 3. This male received Basal IV, (screw-pressed cottonseed meal) + 12% soybean oil meal + Aurofac-Bacitracin + .187% L-lysine; weight 785 grams. Compare with bird in Figure 1, which received the basal containing animal protein.
- Fig. 4. This bird received Basal IV, (screw-pressed cottonseed meal) + 12% soybean oil meal + Aurofac-Bacitracin, no lysine; weight 710 grams. This bird was of a size comparable to the bird in Figure 3 receiving the basal with lysine added.

PLATE V



Fig. 1



Fig. 2



Fig. 3



Fig. 4

EXPLANATION OF PLATE VI

Males from various lots in Experiment II. Chicks pictured are those nearest the lot mean weight at eight weeks.

- Fig. 1. The male in this figure received Basal VI, (screw-pressed cottonseed meal) + 2% meat and bone scraps, 3% fish meal, 10% soybean oil meal and Aurofac-Bacitracin; weight, 914 grams. Note the appearance and size of this bird when compared with that of birds in Figures 1, 2, and 3. of Plate V. The weight of this bird is greater than that of the male on the right in Figure 2. which received the K. S. C. High Efficiency broiler diet.
- Fig. 2. This male received Basal II + Aurofac-Bacitracin. (K. S. C. High Efficiency ration) and weighed 906 grams.
- Fig. 3. This bird received Basal VI (screw-pressed cottonseed meal) + 2% meat and bone scraps, 3% fish meal, 10% soybean oil meal and Aurofac-Bacitracin; heaviest male of all lots; weight, 1181 grams.
- Fig. 4. This bird also received Basal VI, and was the heaviest female of all lots; weight, 922 grams.

PLATE VI



Fig. 1



Fig. 2



Fig. 3



Fig. 4

DISCUSSION

As was stated in the Review of Literature, comparatively few studies have been conducted to study chick growth responses stimulated by supplementation of cottonseed meal diets with specific antibiotics and B₁₂ feeding supplements. More attention has been directed to determining the nutritional value of soybean vegetable protein diets supplemented with growth stimulating feeding supplements. Several studies have indicated that the nutritional value of cottonseed meal is inferior to that of soybean meal, when used as a single source of protein in all-vegetable protein diets. In designing Experiment I the writer attempted to (1) confirm the results that cottonseed meal is inferior in nutritional value to soybean meal when used with antibiotics and vitamin B₁₂ as the sole source of protein in an all-vegetable protein diet; (2) obtain additional information regarding growth responses promoted by the addition of identical levels of specific antibiotic and B₁₂ feeding supplements to a cottonseed meal diet.

To accomplish the above objectives, the two basal diets, I and II, were adjusted so that the calculated vitamin, mineral, amino acid and protein content of each basal was comparable to the recommended nutrient allowances for poultry set forth by the National Research Council (1950). The protein content of Basals I and II was 19.88 and 19.71, respectively.

According to the literature, cottonseed meal was suspected of being deficient in the amino acid, lysine, consequently, all diets containing cottonseed meal in Experiment I were supplemented with .2 per cent L-lysine, (monohydrochloride anhydrous).

All antibiotic and B₁₂ feeding supplements were added at an identical level of 10 grams of antibiotics and 6 milligrams of B₁₂, per ton of feed. The concentration of the antibiotics and vitamin B₁₂ was calculated for each product, and adjusted so that each was supplemented at the previously mentioned level. This level was chosen as a practical and optimum level of supplementation as a result of experiments conducted by Sanford (1952) at the Kansas State Agricultural Experiment Station. The concentration of antibiotics and B₁₂ supplements was not sufficient to attain a level of 10 grams when used alone, thus it was necessary to include crystalline aureomycin HCl, Bacitracin (5 grams of antibiotic per pound of supplement) and Bi-Con TM-5 (5 grams of Terramycin per pound of supplement) in the proper proportion to attain this level.

It was observed that growth of chicks fed the cottonseed meal basals was inferior at all ages to those receiving the soybean meal diets. An analysis of variance of eight week chick weights revealed highly significant differences in growth of the ten lots of chicks. This supports the findings

of Richardson and Blaylock (1950b) and Sherwood and Couch (1950). Results of this study also revealed that although chicks fed unsupplemented cottonseed meal basals were inferior in growth, and grew at a slower rate, the birds were more uniform than those receiving the unsupplemented soybean meal basal as shown by the relatively low standard error of the mean for the lot weights.

As is shown by an analysis of variance, there were highly significant differences in the growth of chicks in lots receiving antibiotics and B₁₂, and those in lots receiving the non-supplemented basals. This is in agreement with the findings previously reported by Newell et al. (1947), Richardson and Blaylock (1950b), Stokstad and Jukes (1950b), Berg et al. (1950), McGinnis and associates (1950), Lawrence et al. (1950) and Johnson (1951).

Under the conditions of this study, it was found that few significant growth differences between lots, resulted from the addition of different antibiotic and B₁₂ supplements to soybean and cottonseed meal basals. This result coincides, to a degree, with the findings of Matterson et al. (1951) regarding their work with soybean all-vegetable protein basals. However, in this study, the combination, Aurofac-Bacitracin appeared to be the most consistent of all antibiotics used, in its growth stimulating properties, when added to the

cottonseed meal diet. Davis and Briggs (1951) combined (aureomycin HCl and streptomycin) and (procaine penicillin G. and bacitracin). The authors cited did not report any evidence of superior growth as a result of these combinations with soybean meal basals. There is a slight possibility that the combination observed in this study may be better adapted to cottonseed meal diets when used as a mixture. Growth was too variable within lots and responses varied too greatly with regard to sex for a definite sequential evaluation of the different antibiotics under observation.

Feed efficiencies of both basals were considerably improved when antibiotics and B₁₂ were added. This result is in agreement with the findings observed by Abbott (1951), Thayer et al. (1950), Davis and Briggs (1951) and Matterson et al (1951). Diets of both basals supplemented with Aurofac-Bacitracin combination were slightly less efficient than other diets. It is the opinion of the writer, that this was due to the additional requirement of feed for maintenance of the heavier birds found in this lot.

There were no positive indications that antibiotics and B₁₂ attributed to the very low mortality rate, observed under the conditions of this study.

Experiment II was designed to determine the extent that cottonseed meal could be used in practical chick diets to promote optimum chick growth. To accomplish this objective, in view of the lower nutritional value of cottonseed meal for chicks as compared with soybean meal, the writer obtained a sample of experimentally processed screw-pressed meal for use in the diets. According to Altschul this cottonseed meal has been reported to be of high nutritional value by several research workers who have tested the meal with chicks.¹ The condition under which this meal was processed, the moisture content, and the gossypol analysis were presented in Materials and Methods.

As was previously stated, cottonseed meal was suspected of being deficient in lysine, thus the writer felt the need of supplementation for more accurate evaluation. In contrast to the level of .2 per cent L-lysine as used in Experiment I, and upon recommendation of the Southern Regional Research Laboratory, the meal was supplemented at an increased level of .308 per cent, or 4.25 grams, per pound of cottonseed meal used in the diet. For critical comparison, Basal II was supplemented with L-lysine in the same manner. In Basal IV, where 20 per cent of the diet was cottonseed meal, and 12 per

¹Personal communication: Ltr. from Dr. A. M. Altschul, Head, Southern Regional Research Laboratory, New Orleans, Louisiana.

cent soybean meal, the level of .187 per cent L-lysine was used. No lysine was added to the basals containing animal protein.

The protein content of Basal IV was 19.92 per cent. Basals V and VI each contained 19.87 per cent protein.

The riboflavin supplement of all basals containing cottonseed meal, was increased from 1.76 milligrams per pound of diet as in Experiment I, to 3.52 milligrams per pound of diet. This increase in riboflavin content was made because of a "curled toe" condition noted in several birds which received basals containing cottonseed meal. Grau (1950) reported such a condition in his investigation with cottonseed meal when the product was included at levels above 30 per cent in chick diets. The author cited, indicated that the requirements for riboflavin as recommended by the National Research Council appeared to be low when cottonseed meal diets are included in diets at high levels.

As shown in Table 2, the vitamin D supplement was reduced to 25 grams (500 A.O.A.C units or 375 I.C.U.'s). This change was made to conform with the recent standard International Chick Unit (75 I.C.U.'s equivalent to 100 A.O.A.C. units). Table 2 also reveals a decrease in vitamin A supplement, added at a level of 40 grams as compared with 100 grams in Experiment I. This was due to an increase in the potency of the new pro-

duct used. The product used in Experiment I contained only 2400 U. S. P. units per gram of supplement as compared with 5000 U. S. P. units per gram in the new product. Other nutrients were adjusted and supplemented as previously described in Experiment I.

In view of findings in the previous experiment, the combination antibiotic and B₁₂ feeding supplement Aurofac-Bacitracin was used, and added to the basals at the same level as in Experiment I.

Further confirmation of the growth stimulating properties of antibiotic and B₁₂ supplements was obtained as a result of this experiment, when it was observed that birds fed basals supplemented with antibiotics and B₁₂ maintained superior growth as compared with those receiving unsupplemented basals. As is revealed by the analysis of variance in Table 2, the antibiotic and B₁₂ supplementation caused a highly significant difference in growth of the birds. Table 9 also presents, highly significant data with regard to variations due to type of proteins used, which confirms the findings of Sherwood and Couch (1950), Richardson and Blaylock (1950b) and Machlin et al. (1952). The authors cited supplemented both soybean and cottonseed meal basals with antibiotics and B₁₂. The results indicated the superior nutritional value of soybean meal when the two were compared.

It was observed in this study that the basal containing screw-pressed cottonseed meal supplemented with antibiotics

and B₁₂ resulted in superior growth as compared with the basal containing the hydraulic cottonseed meal supplemented in the same manner. The difference in the lot mean weights of the birds fed supplemented Basal III and those fed supplemented Basal II was 109.11 ± 29.82 grams, a highly significant difference. These results indicate, that when both types of meal were supplemented, the experimentally processed product was of higher nutritional value. When the types of meal were compared without supplementation, no significant difference was noted between the lot mean weights of the birds receiving both basals. On the other hand, when the lots of birds fed each basal were compared according to sex, a significant difference of 75.57 ± 29.56 grams was noted between the females of both lots compared. The margin was in favor of the birds fed Basal III containing the screw-pressed meal.

The mean weight of birds in Lot 8, (Basal IV, .187 per cent lysine, 12 per cent soybean meal, Aurofac-Bacitracin) was compared with chicks receiving Basal IV, Lot 7, (lysine omitted). The resulting difference was nonsignificant.

When the lot mean weight of birds in Lot 8 was compared with that of birds in Lot 6 receiving Basal III with antibiotics added, and no soybean meal included, similar results were found. The birds in Lot 6, showed a nonsignificant increase in weight as compared with the birds in Lot 7 receiving Basal IV with antibiotics, soybean meal and no lysine. These

findings led the writer to assume that 12 per cent soybean meal adequately replaced the lysine supplementation. Thus making it possible to incorporate soybean meal in a cottonseed meal diet as an adequate source of the deficient amino acid.

As is revealed in Table 11 when the lot mean weight of the birds receiving the diet containing fish meal, plus meat and bone scraps was compared with weights of birds given other basals, the growth produced by these diets was superior to all other diets used, with one exception. This diet was Basal I supplemented with antibiotics. The mean lot weight of the birds in Lot 10 given Basal VI was 909.93 ± 48.01 grams as compared with 906.00 ± 22.61 grams, the lot mean weight of the birds in Lot 4 receiving Basal I supplemented with antibiotics. The small difference of 3.93 ± 53.07 was nonsignificant. When the males and females of each lot were compared, the males of Lot 10 were heavier by a significant difference of 52.42 ± 24.90 grams. Surprisingly, the mean weight of the females in Lot 4 was greater than that of the females in Lot 10; however, the difference of 50.02 ± 42.72 grams was nonsignificant in view of the large standard error of the difference. The previously mentioned results apparently confirm the report by Sherwood and Couch (1950) that cottonseed meal can be incorporated in the diet of chicks and promote

growth equal to that produced by a good soybean meal ration, provided animal protein is added. A further comparison of the birds in Lot 10 fed Basal VI, with birds in Lot 6 fed Basal III supplemented with antibiotics, shows that Lot 10 birds were heavier by 177.64 ± 55.58 , a highly significant difference. This supports the findings of Milligan et al. (1951) whose work indicated that lysine could be omitted from the cottonseed meal diet and satisfactory growth could be obtained, provided an adequate source of animal protein is included. Grau (1950) reported similar conclusions.

The gains made by the birds receiving basals supplemented with antibiotics alone, or with antibiotics and animal protein, were consistently greater than those of the unsupplemented basals as shown by Figs. 7 and 8. The birds in Lot 7 also made greater gains when compared with the birds in Lots 1, 2 and 3. A study of Fig. 8 shows evidence that birds fed diets most adequately supplemented made the most rapid gains after the on-set of Newcastle disease. One exception is the case of birds in Lot 10. These birds gained the least of the birds fed supplemented diets in the sixth and seventh week. In view of the disease, the cause for this result was unknown. The birds in the lots fed unsupplemented basals made the least gains due to their comparative low plane of nutrition.

Results were noted which indicated that, on a lot mean

basis, birds in Lot 9 (Basal V) gave the most favorable feed efficiency. A study of Table 13 reveals that the birds in Lot 10 receiving Basal VI consistently required less feed per gram of gain than all other lots from the first through the fifth week. This indicated that possibly the diet for Lot 10 was utilized as efficiently as that for Lot 9, (Basal V) supplemented with antibiotics and B₁₂. The feed efficiency of the birds in Lot 10 also compared favorably with that of the birds in Lot 4 (Basal I, Aurofac-Escitracin). The birds in Lot 10 had an average efficiency slightly better than those in Lot 4. These results are in agreement with the findings of Machlin et al. (1952) with regard to the effect of dietary antibiotics on feed efficiency. A further study of Table 13 shows that the feed efficiencies of all screw-pressed meal basal diets were more favorable than those of the hydraulic type meal. It appears that the low temperature and moisture conditions under which the meal was processed may have an effect on its nutritional value when compared with the hydraulic product. The comparatively low free gossypol content of the screw-pressed meal may also be a factor supporting its superior nutritive value.

The writer attempted to correlate the effect of antibiotics on the mortality of the birds due to Newcastle. A study of Table 8 shows that the per cent of mortality variations between

lots was too slight to give substantial evidence supporting the influence of antibiotics.

As no symptoms of any nutritional deficiency resulted during the experiment, the writer assumes confirmation of Grau's (1950) hypothesis regarding the higher riboflavin requirements of chicks receiving cottonseed meal.

SUMMARY AND CONCLUSIONS

Experiment I was conducted to (1) compare, in terms of growth, the nutritional value of cottonseed oil meal and soybean oil meal; (2) compare the growth response promoted by supplementing both vegetable sources of protein with identical levels of various antibiotic and vitamin B₁₂ feeding supplements.

Using growth as a criterion, it was found that when cottonseed meal was included in an all-vegetable protein ration at a level of 33 per cent, the product was inferior to soybean meal.

It was observed, that when an all-vegetable protein cottonseed meal diet was supplemented with antibiotics and B₁₂ a highly significant increase in growth was promoted. The addition of antibiotics and B₁₂ feeding supplements to both cottonseed and soybean meal rations at identical levels promoted superior growth in birds fed the soybean basals.

Under the conditions of this experiment, growth was too variable within lots and responses varied too greatly with regard to sex for a definite sequential evaluation of the growth responses promoted by the different antibiotics under observation. The combination supplement, Aurolac-Bacitracin, appeared to be more consistent in its growth stimulating properties than any of the different antibiotics, when added to cottonseed meal diets.

Feed efficiency was observed to be more favorable when antibiotics and B₁₂ were added to the cottonseed and soybean meal diets.

Experiment II was conducted to determine the extent and conditions under which cottonseed meal could be used in practical starting diets to obtain optimum growth of chicks.

Under the conditions of this experiment, it was found that the experimentally processed screw-pressed type meal produced growth in chicks superior to that of the hydraulic type when both were supplemented with antibiotics.

It was observed that an all-vegetable protein cottonseed meal basal diet, supplemented with 12 per cent soybean meal produced growth equivalent to that promoted by the addition of .187 per cent L-lysine. Further observations indicated that the cottonseed meal diet, containing 2 per cent meat and bone scraps, 3 per cent fish meal and the antibiotic-

vitamin B₁₂ feeding supplement produced growth significantly greater than that promoted by an all-vegetable protein cottonseed meal diet, containing 12 per cent soybean meal, and antibiotics with no lysine supplement, as revealed in Table 11.

It was observed in this study that a diet containing 15 per cent cottonseed meal when supplemented with antibiotics, 10 per cent soybean meal, 6 per cent meat and bone scraps, and 6.5 per cent fish meal supported growth comparable to that attained by a practical high efficiency soybean ration.

In view of certain limiting factors encountered while conducting this study, the author realizes that further investigations should be conducted to more thoroughly confirm these findings.

Under the conditions of this investigation, the following conclusions may be drawn as a result of this study:

1. Cottonseed meal is of lower nutritional value than soybean meal when fed as a single source of supplementary protein in chick starting rations.
2. Antibiotic and B₁₂ feeding supplements promote growth when added to a cottonseed meal all-vegetable protein diet.
3. The response promoted by adding antibiotics and B₁₂ feeding supplements to a cottonseed meal all-vegetable protein diet does not produce growth equivalent to that of soybean meal

diets supplemented in an identical manner.

4. The combination Aureofec-Bacitraecin antibiotic and B₁₂ feeding supplements appeared to produce results superior to the other antibiotic supplements used when added to a cottonseed meal all-vegetable protein diet.

5. The incorporation of 12 per cent soybean meal in an all-vegetable protein cottonseed meal diet may be used to adequately compensate for the lysine deficiency in cottonseed meal.

6. Cottonseed meal can be used satisfactorily at a level of 15 per cent in practical starting diets when adequately supplemented with a source of animal protein, soybean meal, antibiotics and vitamin B₁₂.

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ADAPTATION OF ANTIBIOTIC AND VITAMIN B₁₂
FEEDING SUPPLEMENTS TO CHICK DIETS

by

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Two experiments were conducted in an effort to accomplish the following objectives: (1) Using growth as a criterion, compare the nutritional value of cottonseed oil meal and soybean oil meal; (2) compare the growth response promoted by supplementing both vegetable sources of protein with identical levels of various antibiotic and vitamin B₁₂ feeding supplements; (3) determine the extent and conditions under which cottonseed meal can be used in practical starting diets to obtain optimum growth of chicks.

Chicks were battery reared and normal husbandry practices were applied. The Kansas State College High Efficiency ration, minus antibiotic and vitamin B₁₂ feeding supplements, was used as Basal I. All other diets containing cottonseed meal were modified from this basal. The protein, vitamin, mineral, and amino acid content of each diet was adjusted to conform with the National Research Council's Recommended Nutrient Allowances for Poultry.

Antibiotic and B₁₂ feeding supplements were supplemented at the rate of 10 grams of antibiotics and 6 milligrams of vitamin B₁₂ per ton of feed. The antibiotic and B₁₂ feeding supplements used in Experiment I were as follows: Aurofac, (1.8 grams of Aureomycin + 1.8 milligrams of vitamin B₁₂ per pound of supplement), Becitreacin, 3-3 (3 grams of bacitracin - 3 milligrams of vitamin B₁₂ per pound of supplement), Bi-Con 3+3 (3 grams of terramycin + 3 milligrams of vitamin B₁₂ per pound of supplement), and a combination of Aurofac and Becitreacin.

The Aurolac (1.8 + 1.8) - Bacitracin (3 - 3), combination was used to supplement the diets in Experiment II.

An experimentally processed screw-pressed type cottonseed meal obtained from the Southern Regional Research Laboratory in New Orleans, Louisiana was used in Experiment II.

Growth of chicks, which received an all-vegetable protein ration with 33 per cent cottonseed meal was inferior to that produced by a soybean meal ration. When an all-vegetable protein cottonseed meal diet was supplemented with antibiotics and B₁₂, a highly significant increase in growth was promoted. The addition of antibiotics and B₁₂ feeding supplements to both cottonseed and soybean meal rations at identical levels promoted superior growth in birds fed the soybean basal.

Under the condition of this investigation, growth was too variable within lots and responses too varied with regard to sex for a definite sequential evaluation of the growth responses promoted by the different antibiotics under observation. The combination supplement, Aurolac (1.8 + 1.8) - Bacitracin (3-3), appeared to be more consistent in its growth stimulating properties than any of the different antibiotics when added to cottonseed meal diets. The birds fed this combination attained the greatest mean weight of all birds in lots fed supplemented basals.

Feed efficiency was observed to be more favorable when antibiotics and B₁₂ were added to both basals.

Under the conditions of the study conducted in Experiment II, it was found that the experimentally processed type meal produced growth in chicks superior to that promoted by the hydraulic type when both were supplemented with antibiotics and B₁₂.

It was observed that an all-vegetable protein cottonseed meal basal diet, supplemented with 12 per cent soybean meal produced growth equivalent to that promoted by the addition of .187 per cent L-lysine. Further observations indicated that the diet supplemented with cottonseed meal, 2 per cent meat and bone scraps, 3 per cent fish meal and the antibiotic-vitamin B₁₂ feeding supplement produced growth significantly greater than that promoted by an all-vegetable protein cottonseed meal diet containing 12 per cent soybean meal, antibiotics and B₁₂ with no lysine supplement.

The diet containing 15 per cent cottonseed meal, when supplemented with antibiotics and B₁₂, 10 per cent soybean meal, 6 per cent meat and bone scraps, and 6.5 per cent fish meal supported growth comparable to that attained by feeding a practical high efficiency soybean meal ration.