A COMPARISON OF FEEDING HEATED AND NON HEATED PINTO BEAN MEAL TO BROILER-STRAIN CHICKS

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INTRODUCTION

Ever since pinto beans became one of the important dry farm crops, there has been increased interest in the most profitable method of utilizing the cull beans. After threshing, there is always a considerable percentage of beans that are split or otherwise defective, and during good crop years these are rather an important item. The split beans are just as nutritious as those not damaged in threshing, but on account of their appearance they bring but a small price on the market.

Many of the plant proteins can be and are included in poultry diets. Soybean meal has become the one of first choice. Values of other protein concentrates are commonly compared on the basis of substitution of various percentages of soybean meal.

Feed represents the largest single item in the cost of producing broilers. The rate of growth of chicks together with the amount of feed required per unit of gain are the prime criteria by which feed quality is judged and cost estimated. Additional criteria are the quality of feathering, as well as the color and vigor of the birds. There has been, therefore, great interest in all the nutritional findings that affect the growth of chicks.

There is a frequent temptation to employ, in mashes for poultry, some of the cheaper feedstuffs, assuming that since they may seem to have some value for other forms of livestock, they may be used to advantage in poultry feeds.

No data were found in the literature as to the effect of feeding pinto beans to chickens. Therefore, experiments were

initiated to find out a comparison of feeding heated and non heated pinto bean meal to broiler strain chicks.

ORIGIN AND HISTORY

The name "Pinto," as applied to this bean, however, is of rather recent date. It was formerly called either "Mexican" or, in New Mexico, the "Rosillo" bean.

It is not known just why the name Pinto was chosen.

"Rosillo," as used in this connection, conveys the idea of being splashed, having the darker markings irregular in shape and size. The word "Pinto" is commonly understood to mean spotted, having the dividing lines between the colors well marked and very distinct. The bean itself is flecked or splashed, usually having a light background with darker and irregular markings. The background varies in color from a cream or buff to a slightly pinkish tan. On aging, it darkens to a deeper pinkish tan. The markings are olive drab in color and vary from small splotches to narrow bands that run lengthwise of the bean.

The origin of the pinto bean is not definitely known. It belongs to the botanical species "Phaseolus vulgaris," or the common kidney bean (Freeman, 1912; Gracia, 1917). By the French it is known as Haricot and by the Spanish as Frijole. Beans (Phaseolus vulgaris) are thought to have originated in tropical America. They were probably introduced into the southwest by the early Spanish missionaries.

The Phaseoleae, with 47 genera, includes among them the genus Phaseolus as the largest one, with 150 species. The Phaseolus genus will cover the common beans, and the Glycine genus such important members as the soybean.

ECONOMIC IMPORTANCE

Increased production and consumption of beans in the United States have accompanied every major war. The table below shows the United States production of pinto beans, on a cleaned basis, from 1949-62 (Simmons, 1963).

| Year | Production 1,000 cwt | Year | Production 1,000 cwt |
|------|-------------------------|------|-------------------------|
| 1949 | 3,857 | 1956 | 3,351 |
| 1950 | 3,630 | 1957 | 4,913 |
| 1951 | 2.980 | 1958 | 4,904 |
| 1952 | 3,143 | 1959 | 4,381 |
| 1953 | 4.782 | 1960 | 4,475 |
| 1954 | 4.537 | 1961 | 5,592 |
| 1955 | 3,589 | 1962 | 4,105 |

Lantz et al. (1962) conducted research at the New Mexico Agricultural Experiment Station on "Effects of planting date on the composition and cooking quality of pinto beans." They observed that protein contents of the beans from late plantings were slightly higher than those from early plantings. Planting date did not affect the calcium or phosphorus content. The average composition of the pinto bean is:

| Crude protein 2 | 2.7 | Crude fiber | · · · · · · · | . 4.5 |
|-------------------------|------|-------------|---------------|--------|
| Nitrogen free extract 5 | | Fat | | . 1.2 |
| Total protein 2 | 22.7 | Ash | | |
| T.D.N 3 | 56.2 | Digestible | | 9.76 |
| Metabolizable energy | | Productive | | |
| 667 cal | ./lb | | 478 | cal/lb |

Differences in thiamine, riboflavin and niacin were significant.

The nine amino acids that were determined in the beans from early and late plantings were: arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine and valine. Only three amino acids were significantly affected by planting date, when the values were expressed as milligrams per gram of bean (dry basis). They were histidine, methionine and threonine. Only histidine and methionine showed significant differences when the values were expressed as a percentage of total protein.

The shift in the relative amounts of amino acids in beans grown under different environmental conditions may have nutritional significance. As with other plant foods, beans are unbalanced with respect to their content of amino acids and are particularly low in methionine.

REVIEW OF LITERATURE

Like soybean oil meal, the nutritive value of many other legumes is also improved by proper heat treatment. The wide-spread distribution of a trypsin inhibitor in legumes provides the most likely explanation for the observation that heating increases the in vitro digestibility of a number of legumes (Waterman and Jones, 1921; Waterman and Johns, 1921). Jaffe (1950) observed that legumes which had the highest trypsin inhibitor activity were also those in which the digestibility, as measured in vivo, was most improved by cooking. It has been generally noted that supplementation of uncooked legumes with

cystine or methionine markedly improved their nutritive value (Johns and Finks, 1920; Russell et al., 1946; Richardson, 1948; Sherwood et al., 1954). This effect is readily understandable in view of the fact that methionine is the limiting amino acid of most leguminous proteins (Jaffe, 1949a), and hence the action of a proteolytic inhibitor would be expected to accentuate this deficiency in accordance with the view expressed by Almquist (1951).

Several lines of evidence, however, suggest that poor nutritive value of some uncooked legumes cannot be satisfactorily explained by this concept. Klose et al. (1948) found that lima bean fractions possessing high antitryptic activity in vitro also inhibited the growth of rats fed acid hydrolyzed casein. There is no obvious correlation between the effect of heat on the nutritive value of various legumes and the presence or absence of a trypsin inhibitor, a conclusion which also has been reached by Borchers and Ackerson (1950) and Jaffe (1951a).

Hemagglutinins are found in practically all legumes. The possible presence of other growth inhibitors in legumes should not be overlooked. Jaffe (1949b and 1951b), for instance, has postulated the existence of a heat-labile toxic factor in kidney beans and claims to have isolated a fraction which is markedly toxic at a dietary level of 0.05 percent.

Contrary to the beneficial effects of heat observed with most legumes, the nutritive value of the field pea is damaged by baking, canning or autoclaving (Everson and Heckert. 1944:

Richardson, 1948; Murray, 1948; Armbruster and Murray, 1951). This impairment in nutritive value is amenable to correction by supplementation with cystine or methionine (Woods et al., 1943; Schneider and Miller, 1954). Since methionine is the limiting amino acid of pea protein, it would appear that destruction of cystine is the primary factor leading to the methionine deficiency of cooked peas.

Johns and Finks (1920) and Fink and Johns (1921) studied the effect of cooking on the nutritive value of the globulin-like protein of the navy bean and the velvet bean, designated as Phaseolin and Stizolobin, respectively. Cooking was found to improve the growth-promoting properties of these isolated proteins, particularly when supplemented with cystine. In vitro digestibility studies on Stizolobin conducted by Waterman and Jones (1921) indicated that cooking likewise enhanced the susceptibility of this protein to the combined action of pepsin and trypsin.

The beneficial effect which proper heat treatment exerts on the nutritive value of some vegetable protein meals is related to the concomitant inactivation of specific heat-labile factors which elicit deleterious physiological responses in animals. In some cases (for example the trypsin inhibitor and hemagglutinin of soybeans, free gossypol of the cottonseed, and goitrogenic factor of rapeseed) these factors have been well characterized, but in many instances little is known about them except they do exist.

The impairment in nutritive value which follows the excessive application of heat is associated with certain profound changes in the protein molecules themselves or changes resulting from the interaction of the protein with carbohydrate-like components which accompany protein in crude meals. In either event these modified proteins become more refractory to enzymatic (trypsin) attack with a consequent retardation in the rate with which the essential amino acids are released during digestion. The formation of peptide linkages which cannot be split by trypsin may arise in two ways: (1) the interaction of the non peptide carboxyl groups of glutamic and aspartic acids with the non peptide amino groups of lysine or arginine giving rise to atypical peptide linkage, and (2) the interaction of the non peptide amino group of lysine and arginine with reducing sugars so modifying the substrate that it no longer conforms to the specificity requirements of trypsin.

In the animal organism, the decreased protein digestibility induced by heat may lead to the excretion of substantial portions of the protein, thus depriving the animal of the amino acids contained therein if one of these amino acids is limiting in the original protein, the animal will suffer from a deficiency of this particular nutrient. Thus, soybean oil meal, which is suboptimal with respect to methionine, becomes critically deficient in this amino acid when overheated, and, similarly, excessive heat treatment causes the marginal level of lysine in cereal products and cottonseed meal to become critical deficiencies.

The destruction of lysine further accentuates the critical need for this amino acid and, in certain instances, may even precipitate a lysine deficiency with proteins which are normally considered fairly good sources of lysine. The denaturation of protein by heat is characterized by lessened solubility in aqueous solvents.

Soybeans

Shortly after the soybean was introduced into America as a commercial crop, Osborne and Mendel (1917) conducted a study of its potential value as a source of protein for animals, and observed that soybeans would not support the growth of rats unless they had been cooked for three hours. The literature now abounds with innumerable reports confirming the superiority of heat-processed soybean oil meal, not only for rats but also for mice, chicks, turkey poults, swine and human beings.

In general, these studies have shown that the degree of improvement in nutritive value effected by heat treatment is dependent on temperature, duration of heating, and moisture conditions. The data of Hayward et al. (1936) and Wilgus et al. (1936) illustrate the extent to which the nutritive value of soybean protein may be improved, depending on the conditions of processing. Two additional points are evident from these data, namely, the ineffectiveness of dry heat, and the marked impairment in nutritive value which accompanies excessive heat treatment.

In accordance with the general belief that the biological value of a protein is determined largely by its amino acid content, studies have been undertaken by Block and Mitchell (1946) and Oser (1951) to determine if supplementation of the unheated protein with various amino acids would achieve the same effect as heating. It has thus been well established that the addition of methionine or cystine to unheated soybean meal improves protein utilization to essentially the same extent as proper heating (Hayward and Hafner, 1941; Almquist et al., 1942; Clandini et al., 1946; Evans and McGinnis, 1946; Russel et al., 1946; McGinnis and Evans, 1947; Evans et al., 1951). Amino acid analysis, however, revealed the methionine content was substantially unchanged by the amount of heat necessary to produce maximum nutritive value (Almquist et al., 1942; Melnick et al., 1946; Riesen et al., 1947; Evans and McGinnis, 1948; Ingram et al., 1949; Kuiken and Lyman, 1949). Although heated soybean meal was somewhat more digestible than the unheated meal in experiments conducted with rats, the difference was too small to account for the marked differences in biological value (Hayward et al., 1936; Mitchell et al., 1945; Melnick et al., 1946; Carroll et al., 1952). In fact, the absorption of nitrogen, sulfur or methionine itself from the digestive tract of the rat was essentially the same for both the raw and heated protein (Johnson et al., 1939; Melnick et al., 1946; Carroll et al., 1952; Liner and Wada, 1953). A comparison of the absorption of nitrogen from the terminal 20 percent of the small intestine, however, revealed that over twice

as much nitrogen was absorbed from the heated protein as from the raw (Carroll et al., 1952). Failure to confirm this observation has been reported (Borchers, 1953). Regardless of the site of the absorption of the bulk of the protein, the retention of nitrogen and sulfur from raw soybean protein is nevertheless significantly less than from the heated protein (Johnson et al., 1939). From these observations, it would appear that the lower nutritive value of unheated soybean meal was not the result of incomplete digestion of the protein, but rather that the methionine was absorbed in a form, or possibly at a site, from which it cannot be effectively utilized for growth.

Trypsin Inhibitor. Melnick et al. (1946), on the basis of observations on the in vitro release of amino acids from soybean protein by the enzyme pancreatin, suggested that methionine of raw soybean protein was liberated more slowly by the proteolytic enzymes of the intestines than the other essential amino acids so that it was not available for mutual supplementation of the latter. This concept was supported by the discovery, purification and characterization of a heat labile protein in soybeans which inhibits the proteolytic activity of trypsin (Bowman, 1944; Ham and Sandstedt, 1944; Kunitz, 1945, 1946; Bowman, 1946, 1948, 1950; Borchers et al., 1947; Kunitz, 1947a, 1947b). Active antitryptic preparations from unheated soybeans have been shown to retard the growth of rats (Klose et al., 1946; Borchers et al., 1948; Liener et al., 1949), mice (Westfall and Hauge, 1948) and chicks (Ham et al., 1945). Because the protein efficiency of

partially heated soybean flour increased in proportion to the destruction of the trypsin inhibitor, Westfall and Hauge (1948) concluded that the trypsin inhibitor was the major cause of poor utilization of the protein in the raw soybeans. Almquist and Merritt (1952a,b) found the growth inhibition of chicks was almost fully developed when as little as one fourth of the dietary protein was furnished in the form of the raw meal. Crude or crystalline trypsin was capable of reversing this growth inhibition (Almquist and Merritt, 1953a). Cornell workers. Nesheim et al. (1962), reported a slight growth depression when chicks were fed a crystalline soybean trypsin inhibitor, but the depression was not nearly as great as that obtained with 15 percent raw soybean meal. Fractionation studies showed that fractions which contain no detectable trypsin inhibitor markedly depressed growth (Saxena et al., 1963). The evidence indicates that although trypsin inhibitors in raw soybeans may have some physiological effect on birds, they apparently do not account for the major part of the growth depression.

The foregoing evidence strongly suggests that the poor growth promoting qualities of raw soybeans can be attributed to an inhibition of intestinal proteolysis. Almquist and Merritt (1951 and 1953b) believed that action of the inhibitor is a general interference with digestion so that a substantial amount of the most limiting amino acid of soybean protein, methionine is excreted unabsorbed, thus precipitating a methionine deficiency (Almquist et al., 1942). In confirmation of this concept, these

authors have shown with chicks that the addition of the trypsin inhibitor in the form of raw soybean meal to rations containing marginal levels of lysine, arginine, isoleucine or tryptophan caused these rations to become markedly deficient in these particular amino acids.

Hemagglutinin. Many legume species contain substances that can agglutinate the red blood cells of animals. Liener (1953) purified a hemagglutinin found in raw soybeans and showed that this substance (once called soyin) depressed growth of rats. However, it was added at a very high level when considered in terms of the quantity of raw soybean meal used as a starting material for the fractionation. It is probable that hemagglutinin does not account for the marked growth depression obtained with levels of raw soybean ordinarily used in experimental diets. Recently, black beans and kidney beans have been shown to contain potent hemagglutinin, and when fed to rats at lower levels they markedly depressed growth, and at high levels they caused 100 percent mortality (Honaver et al., 1962).

Amino Acid Supplementation. Hayward et al. (1936) showed that supplementing raw soybean diets for rats with methionine and cystine markedly improved protein efficiency ratios. Work in the 1940's further suggested that unavailability of amino acids was a major factor in explaining the growth inhibition of raw soybean meal. As a result, a number of studies have been reported on attempts to counteract the growth depression by supplementing diets with free amino acids.

Fisher and Johnson (1958) overcame nearly all the growth depression in chicks by supplementing a raw soybean meal diet with a mixture of essential amino acids. Borchers (1961) and Booth et al. (1960) confirmed this observation with rats, using only four amino acids. However, the latter workers failed to supplement the heated soybean control diet with methionine and the other essential amino acids used even though it is recognized that the diet they fed was deficient in sulfur amino acid.

Saxena et al. (1962a) showed that although part of the growth depression could be counteracted by adding four or more essential amino acids to the diet, it was not possible to completely counteract the growth depression. Furthermore, the pancreatic hypertrophy and poor food utilization observed in chicks fed raw soybeans were not corrected. Barnes et al. (1962) also were not able to correct the growth depression in rats by amino acid supplementation. It would, therefore, appear that growth depression by raw soybeans is not due simply to the unavailability of certain essential amino acids.

Antibiotics. Borchers et al. (1957) reported that growth depression in rats fed raw soybean meal could be prevented by including high levels (0.1 percent) of penicillin and streptomycin. Inclusion of only one antibiotic did not greatly influence growth rate. An extension of these studies to the turkey poult (Lenerode et al., 1961) showed that antibiotics were effective in reversing growth depression only when low levels of raw soybeans were used. At higher levels, growth rate was somewhat

improved by the antibiotics, but the growth rate did not come near approaching that obtained with heated soybeans. The latter workers also showed that adding 0.25 percent of dietary enzymes failed to influence the growth rate of turkeys fed diets containing raw soybeans. Zinc supplementation was ineffective.

Fat Absorption. Workers at Cornell have observed that fat absorption is markedly reduced in young chicks by including raw soybean in the diet (Nesheim et al., 1962). This defect occurs only during the first two weeks of the chick's life. Adding trypsin partially counteracted the raw soybean effect. Although this is an interesting observation, it still does not appear to explain the growth inhibiting properties of raw soybean meal. Growth in chicks after two weeks of age is depressed even though fat absorption is no longer affected.

Physiological Changes. The feeding of unheated soybeans to chicks or rats markedly increases the size of the pancreas in relation to body weight (Chernick et al., 1948). In chicks, the weight of the pancreas per unit of body weight will be about double that of chicks fed a normal ration. This effect is specifically associated with some factor in the raw soybean, and not simply caused by the unavailability of certain essential amino acids. Development of amino acid deficiency by the use of other dietary components does not change the pancreas size. Furthermore, adding amino acid mixtures to diets containing raw soybeans did not prevent the pancreatic hypertrophy (Booth et al., 1960; Saxena et al., 1962a).

It has been proposed that depression in growth is related to a depletion of essential amino acids caused by a continuous excessive secretion of pancreatic enzymes. This idea was supported by evidence showing higher levels of certain pancreatic enzymes in the pancreas as well as in the feces of birds fed raw beans (Lepkovsky et al., 1959). The total amount of nitrogen lost through excess secretion of pancreatic enzymes would not seem large enough to account for the marked growth depression observed in animals fed raw soybeans. Furthermore, supplementing diets with high levels of essential amino acids or additional dietary protein did not completely overcome the growth depression (Barnes et al., 1962; Saxena et al., 1962b).

A significant recent observation (Saxena et al., 1963) suggested that the mechanism for release of pancreatic enzymes into the digestive tract is affected by some factor in raw soybeans and that the hypertrophied pancreas may be caused by accumulation of pro-enzyme material. In normal animals, injecting the drug Pilocarpine (which stimulates the parasympathetic nervous system) will enhance the flow of pancreatic enzymes to the digestive tract. Injection of Pilocarpine significantly reduced the amylase activity in the pancreas of chicks on heated meal, but had no appreciable effect on pancreatic amylase activity of raw meal fed chicks. Furthermore, histological examinations showed an accumulation of Zymogen material in the pancreas of chicks fed raw soybeans and little or no Zymogen material in the pancreas of chicks fed heated soybeans.

Some other physiological changes that have been observed by Saxena et al. (1962c) in chicks fed raw soybeans are decreased glycogen content of the liver and muscle, and an increased oxygen consumption. However, these changes were prevented by adding a mixture of essential amino acids to the raw soybean diet. The free amino acids may have been used for gluconeogenesis which prevented a reduction in glycogen levels. With rats, Inamdar and Sohoni (1961) observed a considerable decrease which occurred in the activity of several enzymes (xanthine oxidase, lactic and succinic dehydrogenases, and glucose 6-phosphatase) in the liver. Levels of glycogen, total SH compounds, riboflavin and flavin adenine dinucleotide were markedly reduced.

Effect of Age. Age of animals has a marked effect on the way they respond to diets containing raw soybeans. Fisher et al. (1957) reported the laying hen was insensitive to inhibitors in raw soybeans. In an experiment involving starting of chicks at different ages on soybean diets, the chicks were found to be insensitive to growth depression after six weeks of age. However, pancreatic hypertrophy was observed at all age groups up to 12 weeks of age, although it was not as marked in the older birds. The hypertrophy was reversible within 72 hours after chicks were changed from raw-to heat-treated soybean meal. Alumot and Nitsan (1961) showed that intestinal proteolysis was completely inhibited in chicks fed raw soybeans up to three weeks of age. Whether or not the proteolytic enzymes observed in the older chicks were from pancreatic secretion or intestinal secretion was not determined.

The popular belief that anti-trypsin factors account for the growth depression has not been supported by experimental evidence. Raw soybeans contain a factor which agglutinates red blood cells and depresses growth in rats when fed at high levels. It appears, however, that the major growth depressing factor in raw soybeans is present in a water-insoluble residue fraction, the nature of which remains to be determined (Jensen and Saxena, 1963).

Velvet Beans

Tempelton et al. (1917) reported that velvet beans were relished by steers, but were unpalatable to dairy cows. Satisfactory results have been reported with the use of velvet beans for fattening hogs. Tempelton et al. (1917) found that feed for fattening pigs could contain 50 percent velvet beans, but results were not as good as those obtained when other sources of protein were used. Scott (1917) reported that satisfactory gains were obtained with pigs when the feed contained shelled corn, velvet beans in the pod and Japanese cane. Later results of Tempelton (1920) indicated that for best performance the feed for pigs should not contain more than one part of velvet beans for each four parts of corn.

Sure and Read (1921) observed that raw velvet beans were injurious to young rats when constituting 40 percent of the total ration. They also reported that autoclaving the seed for one hour at 15 pounds pressure destroyed most of the toxic effect, making it possible to include 60 percent autoclaved beans in the

ration. Read (1923) confirmed the observation that autoclaving of the beans destroyed the toxic factor.

Salmon (1922) reported that velvet beans were toxic to pigeons. He found that on the second day of feeding, birds showed ruffled feathers and appeared sleepy. Mortality commenced on the sixth day, and survivors were in poor condition, having lost about 80 grams.

Harms et al. (1961), working with chicks, observed that a depression in growth rate was noted with chicks fed diets containing 12.5, 25.0, 37.5 and 50 percent velvet beans. This depression in growth was detected at the end of the first week of feeding. An increase in velvet bean content of the diet resulted in a greater depression of growth. It was concluded the poor results were attributed to an unknown toxic factor in velvet beans. Feeding diets containing 50 percent velvet beans caused 25 and 15 percent mortality in Experiments 1 and 2, respectively. Most of this mortality occurred during the first week. Hemorrhage was the principal lesion found on autopsy. These were seen in the thigh musculature and the mucosal surface of the duodenum and esophagus. Histologically, there was edema and hemorrhage in the lamina propria. It is possible that velvet beans could be rendered non toxic for poultry by heat treatment or autoclaving.

Legume Seeds

Borchers and Ackerson (1950) fed ll species of legume seeds, raw and autoclaved, to rats as the sole source of protein at a 12 percent protein level. The jack bean, lentil, velvet bean, horse bean and black eyed pea were improved by autoclaving, while the peanut, partridge pea, guar, lespedeza, mung bean and common vetch were not improved. No correlation was observed between the effect of autoclaving on nutritive value and the presence or absence of the trypsin inhibitor in the raw legume seeds. Kienholz et al. (1960) observed that diets containing raw legume seeds depressed growth of chicks below that of chicks fed a cornsoybean oil meal control diet. Autoclaving of the seeds improved performance of chicks in all instances.

Double Beans (Faba vulgaris)

Sohoni et al. (1958) concluded that so far as the trypsin inhibitor is concerned, Faba vulgaris resembles soy and lima beans, and that like the lima bean it appears to lose its activity when autoclaved.

Horse Beans

Mohon and Common (1950) observed that protein of the horse bean is deficient in methionine and to a smaller extent in cystine. There are also slight deficiencies of tryptophan, glycine, phenylalanine and threonine. Bletner et al. (1963) observed that diets containing 33 percent horse beans and 5 percent fish meal supplemented with either autoclaved soybeans or soybean oil meal, as well as diets containing 60 percent horse beans, 5 percent fish meal, 5 or 10 percent added fat, and the

necessary vitamins, minerals and amino acids produced satisfactory chick growth.

Chick growth was unsatisfactory when birds were fed the 33 percent horse bean--12 percent fish meal diet; the 33 percent horse bean--raw, ground soybean diet; and the 60 percent horse bean--5 percent fish meal diet, with or without 15 percent added fat.

Black Beans

Jaffe et al. (1955) found that raw beans (black beans) reduced rate of growth, total digestibility and digestibility of protein and fat in rats. Food consumption diminished, but not enough to account for reduction of growth rate. In an experimental diet, the addition of only 5 percent raw beans caused loss of weight, but the same amount of cooked beans permitted an increase.

Lima Beans

Richardson (1948) observed that heated lima and pinto beans (autoclaved at 15 pounds pressure for 30 minutes) were superior to the raw beans; heated southern and English peas were not. The addition of 0.2 percent methionine increased the nutritive value of all the heated legumes; the heated English peas with methionine were superior to egg albumen. Methionine improved raw lima beans slightly and raw English peas markedly. In tests with southern peas and pinto beans, 0.2 percent methionine was required to give the maximum rate of growth.

Klose et al. (1948) reported that when the lima bean protein fractions were heated for 30 minutes at 15 pounds steam pressure, the growth rate was equal to that on the basal diet. The growth depressing effect of the bean fractions was equal to that of a diet containing unhydrolyzed protein. It is concluded that lima bean protein fractions, with high trypsin inhibiting activity, exert their growth inhibiting effect by some mechanism not directly related to inhibition of the normal enzymic hydrolysis of dietary protein.

Klose et al. (1949) observed that growth of rats given raw beans was depressed. Growth was improved slightly by the addition of methionine and markedly by heating the beans. A combination of two treatments increased growth approximately to that of control animals.

Similar experiments with crude lima bean protein instead of the whole bean gave similar results. Methionine at the 0.6 percent level counteracted the growth depressing effect of the lima bean protein; at the higher level, methionine was not so effective.

Tauber et al. (1949) found that a fraction designated as "orude trypsin inhibitor" was isolated from lima beans. It was a crystalline product of a globulin nature, which powerfully inhibited the proteolytic action of trypsin. It was exceptionally stable to heat although autoclaving for one and one-half hours at 18-pound pressure destroyed its inhibitory action.

Field Beans

Riley (1961), while studying the nutritional value of cull field beans in chick diets, made the following observations: (1) Chicks maintained on diets containing high levels of raw beans lost weight and approached 100 percent mortality after seven days. (2) Supplementation of bean diets with zinc, methionine, trypsin, threonine and valine had no effect on the growth rate of chicks. (3) Dry autoclaving beans for 60 minutes at 10 p.s.i. or 30 minutes in the presence of 10 or 20 percent water optimumly improved growth. (4) Uric acid determination indicated that excreta from birds fed diets containing autoclaved beans contained considerably less uric acid than excreta from the control diet. (5) The metabolizable energy value of raw and cooked beans was 204 and 499 cal/lb, respectively. This indicated that cooking more than doubled the metabolizable energy of cull field beans. (6) Papain or fungal enzyme supplements did not improve the utilization of raw or cooked beans. (7) Soaking beans in water and discarding the soaking water prior to autoclaving failed to increase the nutritional value of beans. (8) Slight growth improvement was observed when 0.5 percent methionine was added to diets containing cooked beans. (9) Supplemental tallow, higher levels of vitamins and trace minerals did not improve chick growth. (10) Various fractionation methods were unsuccessful in isolating or concentrating the growth depressing factor(s) in raw beans. (11) When beans were treated according to Kunitz's method for the isolation of the trypsin

inhibitor (developed using raw soybean oil meal), the growth depressing factor(s) was not concentrated or isolated. However, this fraction was found to be rich in the trypsin inhibitor. These results show the growth depressing factor(s) in beans was not identical with the trypsin inhibitor.

Navy Beans

Kakde and Evans (1963) observed that rats fed raw bean flour as the sole source of dietary protein lost weight and died. Supplementation of the diet with vitamin B12, methionine or levels of all essential amino acids in an amount required by the rat failed to improve growth rate or protein efficiency. Antibiotic supplementation prevented weight losses and mortality, and in combination with methionine and vitamin B12 produced limited growth. Germination of the bean seeds did not improve their growth promoting value. Rats fed beans that had been heated in the autoclave at 121°C for five minutes gained weight and did not die, and rats fed heated beans supplemented with methionine grew as well as rats fed casein to supply a similar level of protein. Beans which had been heated for longer than five minutes did not promote as good growth as those heated for five minutes. Destruction or inactivation of some of the essential amino acids occurred in overheated beans.

Red Kidney Beans

Wagh $\underline{\text{et}}$ al. (1963) experimented to ascertain the toxicity of red kidney beans when included in the chick diet as a source

of protein. Inclusion of raw beans in the diet at a level of 30 percent or more produced growth retardation and pancreatic hypertrophy. Autoclaving the beans at 120°C for 30 minutes considerably improved the weight gains and eliminated pancreatic enlargement. Supplementing the diet containing 50 percent autoclaved beans with deficient essential amino acids (compared to a corn-scybean diet) did not improve growth performance to the level obtained with the controlled diet. A diet containing 1 percent of a hemagglutinin isolated from raw beans significantly depressed growth rate without pancreatic enlargement. The latter observation was of interest since the growth depression caused by a trypsin inhibitor of legume was accompanied by pancreatic enlargement, and points to the multiplicity of a toxic effect obtained with legumes.

The growth performance of chicks fed the autoclaved kidney beans was subnormal compared with the control diet, and an attempt was made to determine whether differences in the dietary amino acid content might explain this effect. The results show that equating the kidney bean diet with that of the control in terms of amino acid composition did not eliminate the growth differences. Thus, the unresolved growth depression noted with the autoclaved diet containing red kidney beans appears to be due to other factors such as thermostable toxic principle, poor availability of amino acids other than those added or a dietary energy deficiency.

Pinto Beans

Wilson and Lantow (1926) reported that raw pinto beans are a satisfactory feed for fattening lambs. The feeding value of these beans is approximately 85 percent that of corn.

Cooked pinto beans are a satisfactory feed for hogs, but the raw beans are unsatisfactory.

Titus (1924) observed that pinto bean culls cannot be fed in amounts greater than four pounds per day, to steers of 500 to 600 pounds live body weight, without causing them to scour. The indications are that pinto bean culls have a toxic action when fed in amounts greater than as above indicated. Cooking seems to destroy the apparent toxicity.

MATERIALS AND METHODS

Two separate experiments were conducted at the Kansas State University poultry farm in the poultry nutrition laboratory. A total of 548 chicks were used in the two experiments. Experiment 1, consisting of 180 chicks, was initiated on January 17, 1963 and was run until March 14, 1963. The second experiment lasted from April 25, 1963 to June 20, 1963 and utilized 368 chicks. Cobb's strain-cross White Rock straight-run, day-old, broiler-strain chicks were used in both experiments. The chicks were randomized into 36 lots of 15 chicks each, except for the two control lots of Experiment 2 where 19 chicks were used per lot. The chicks were wing banded and vaccinated when one-day-old by the intranasal method for Newcastle and bronchitis. The birds were maintained in electrically heated, thermostatically

controlled battery brooders with raised wire floor, in a temperature controlled and ventilated laboratory to four weeks of age, at which time they were transferred to unheated growing batteries until the end of the experimental period. Experimental diets and tap water were supplied ad libitum.

Chicks were individually weighed for each two-week period, and the feed consumed during the period was recorded. The experiment was conducted in replicate pens except for two treatments in Experiment 1.

The composition of the experimental diets and the control diet is shown in Tables 1 and 2. Analysis of the pinto beans indicated that the percentage of protein and fat was 23 and 1.2 percent, respectively. The composition of essential amino acids is shown in Table 3.

All diets were formulated to contain adequate amounts of all nutrients known to be required by the chick, including DL-methionine. Since digestibility coefficients for nutrients are not used for calculating diets for poultry, the experimental diets were formulated on the basis of gross composition of ingredients and do not reflect differences in nutrient availability that might result from heating the raw pinto bean meal. The pinto bean component of the diet was varied to be raw meal, heated meal, beans pelleted and reground, formula pelleted, beans pelleted reground and complete formula pelleted.

Pinto beans were heated by putting them in the conditioning chamber of the California Pellet Mill and heating them with live

¹ All tables appear in the Appendix.

steam. Temperature of the bean, taken at the front and back, was maintained at 100° C for approximately 35 minutes (\pm 2 minutes). Following heat treatment, the beans were put in the horizontal cooler and cooled. As the beans were still warm when removed from the cooler, they were placed on racks and spread out for one hour of additional cooling. Both the heated and the unheated pinto beans were ground through a 1/8-inch hammermill screen.

Experiment 1 was conducted as a preliminary test to determine the influence of different levels of heated and unheated pinto beans and their effect on growth. The experimental diets of Experiment 1 were mixed in the feed building located at the University poultry farm, and their composition is shown in Table 1. The composition of the diets used in Experiment 2 is indicated in Table 2.

The different methods used in processing feed for use in formulating rations to study whether heat required in processing was sufficient to get beneficial effects from the pinto beans were: beans ground and pelleted, formula pelleted, pelleted beans reground and formula pelleted (Table 10).

All of the diets used in the experiment were isonitrogenous. In Experiment 2, the calorie content of the experimental diets G and J were adjusted to the level of the control diet by the addition of 3.0 percent and 5.4 percent stabilized animal fat, respectively, to see whether inclusion of fat had any supplementary effect. The calorie protein ratio in the above diets was 43.5, while in other diets it was not controlled, and ranged from 39.2 to 41.4.

DL-methionine was added to the diets to bring the total level of DL-methionine and cystine in the diets to 0.8 percent, as recommended for chicks.

In Experiment 2, the experimental diets were prepared at the Department of Flour and Feed Milling Industries pilot feed mill.

Analysis of variance techniques used are described by Snedecor (1956). Analysis of variance was run on eight-week weight gains and feed conversion data for each experiment. Treatment means were compared at 0.05 percent level of probability, by using Fisher's L.S.D. method as described by Snedecor (1956). The treatment means underscored by the same line are not significant different (P<0.05) from one another.

RESULTS

Experiment 1

The effect of different dietary levels of heated and unheated pinto beans on growth is shown in Table 9.

An analysis of variance was run on the eight-week weight gains. The analysis indicated that diets were significantly different (Table 5). The analysis of feed conversion data showed that diets were nonsignificant at the 5 percent level. However, the F-ratio (see Table 7) approached significance at the 10 percent level of probability.

Chicks fed diets containing either 7.5 percent unheated or 7.5 percent or 15 percent heated pinto beans did not gain

significantly different (P<0.05) than the corn-sorghum grain-soybean oil meal control.

The weight gain of chicks fed diets containing either 7.5 percent heated or 7.5 percent unheated; 15 percent heated or 15 percent unheated pinto beans was not significantly different from each other.

No variable differences in weight gain were observed when chicks were fed rations containing 7.5 percent or 15 percent unheated or 52 percent heated pinto beans.

When the unheated pinto bean level was increased to 52 percent in the chick rations, the weight gain was significantly lower (P<0.05) when compared to all other lots. There was 40 percent mortality, which occurred in the early part of the second week.

The analysis of variance suggested (Table 7) that feed efficiency for chicks fed either the control diet or 7.5 percent heated pinto beans was significantly better (P < 0.10) than diets containing 7.5 percent and 15 percent unheated or 15 percent heated pinto beans.

However, there was no significant difference (P<0.05) in feed efficiency for chicks fed diets containing either 7.5 percent or 15 percent unheated and 15 percent heated pinto beans. When chicks were fed 52 percent heated pinto beans, the feed efficiency appeared to be better than that for those fed unheated pinto beans at the same level. Incorporation of heated pinto beans in the chick ration improved the feed efficiency over that of the unheated ones at the same level.

When heated or raw pinto beans were added to a corn-sorghum grain-soybean oil meal diet, the growth rate gradually decreased. The depression was much more severe for raw beans. These results show that more than 7.5 percent heated or unheated beans were necessary to depress growth.

When the heated pinto bean level was increased from 15 to 52 percent in the chick ration, the growth rate was depressed by 9.7 percent. Similarly, when the level of unheated pinto beans was increased from 15 to 52 percent in the chick ration, the growth was depressed by 32.3 percent. Diets containing raw pinto beans depressed the growth of chicks below that of the cornsorghum grain-soybean oil meal control diet. In other words, growth depressed as the level of the unheated pinto bean increased.

As expected, regardless of the diet, the males grew better than the females within the same treatment. The diet x sex interaction was significant, which showed that the effect of the diet on weight gain had a variable effect from sex to sex.

Experiment 2

The effect of different dietary levels of heated and unheated pinto beans on growth is shown in Table 10. An analysis of variance was run on the eight-week weight gains. The analysis indicated that the effect of the diets on weight gain was significantly different (Table 6).

Although there were small differences in feed efficiency between varying lots, the analysis of variance suggested they were not statistically significant at the 5 percent level of probability (Table 8). Chicks fed diets containing 10 percent unheated pinto beans gained significantly less (P<0.05) than those fed heated pinto beans at the 10 percent level. However, raising the level from 10 to 15 percent of unheated pinto beans did not bring any marked improvement. Similarly, raising the level of 10 to 15 percent of heated pinto beans also did not result in a marked difference. However, there was a slight increase in weight gain when chicks were fed the diet containing 10 percent heated pinto beans. There was a slight but not significant difference in weight gain with 20 percent heated pinto beans, when compared to the group fed unheated pinto beans at the same level. This indicates that the heat treatment of pinto beans probably is more important for better growth than the level of pinto beans in the chick ration.

No appreciable differences in weight gains were noted when chicks were fed rations containing 15 percent ground pinto beans either unheated or pelleted and reground, the formula pelleted, or unheated ground pinto beans pelleted, reground and the complete formula pelleted. The above ration did not significantly differ even from the corn-sorghum grain-soybean oil meal control, although there was a slight increase in the weight gains in the latter.

Addition of fat to either 10 or 20 percent heated pinto beans brought about a small but not significant difference in weight gain when compared to the chicks fed the same diets without fat. As expected, these results were significantly greater than the weight gains obtained by feeding chicks the unheated pinto beans at the same level. This indicates that addition of fat had no significant effect on the weight gain of chicks after the pinto beans had been heated. However, diets containing fat were significantly better (P<0.05) than the cornsorghum grain-soybean oil meal control diet.

Making up the level of DL-methionine and cystine to the standard of 0.8 percent in the chick ration containing unheated pinto beans did not support growth to the level of heated ones having the same standard of DL-methionine. It appears, therefore, that heat treatment to raw pinto beans is essential to remove the growth depressant factor.

As expected, regardless of the diet, the males grew better than the females within the same treatment. The diet x sex interaction was significant, which showed that the effect of the diet on weight gain had a variable effect from sex to sex.

DISCUSSION

In Experiment 1, when heated or unheated pinto beans were added to the chick ration, growth rate markedly decreased when compared to those fed the control diet containing corn-sorghum grain-soybean oil meal (Table 9). The depression in growth rate was more severe with unheated than heated pinto beans. The data also indicated that very high levels; that is, above 7.5 percent or 15 percent heated pinto beans, caused a significant depression in growth rate when compared to the control group.

Almquist (1938) observed that cull pink beans at the 10 percent level apparently were well utilized with no observed detrimental effect. Almquist and Merritt (1952) found that the growth inhibition of chicks was almost fully developed when as little as one fourth of the dietary protein was furnished in the form of the raw soybean oil meal. Riley (1961) observed that more than 10 percent cooked beans were necessary to depress growth, but that only 10 percent raw field beans were necessary to markedly depress growth. The results obtained in Experiment 1 agree, to some extent, with the results obtained by Almquist (1938), Almquist and Merritt (1952) and Riley (1961).

The depression in growth rate in respect to raw pinto beans may be due to the low coefficient of digestibility as was reported by Woods (1952) in respect to field beans as a dietary source of protein for rats. Wagh et al. (1963) concluded that the growth depression noted with the autoclaved diet containing red kidney beans appeared to be due to other factors such as a thermostable toxic principle, poor availability of amino acids or a dietary deficiency.

The results of Experiment 1, however, differed from those of Experiment 2 in that, weight gain of chicks fed diets containing 15 or 20 percent unheated pinto beans did not gain significantly different than the control although there was a slight increase in weight gain in favor of the control diet. Although no satisfactory explanation could be offered for this difference, it is assumed that hot weather may have been a factor which caused a lesser weight gain among all lots in Experiment 2.

The results of Experiments 1 and 2 show that unheated pinto beans in the chick ration from 7.5 to 20 percent did not depress growth when compared to corn-sorghum grain-soybean oil meal control. Wagh et al. (1963) observed that raw red kidney beans in Experiment 1, at a level of 30 percent or more, significantly lowered the growth rate. However, in Experiment 2, the same parameters were affected when the diets contained 15 percent raw red kidney beans. This variation in the experimental results is attributed to the age of the birds; that is, younger birds were more susceptible to the toxicity. The results obtained in this study are in agreement with the above results.

Feeding diets containing 52 percent unheated pinto beans caused 40 percent mortality, which occurred in the early part of the second week. Harms et al. (1961), in his studies with velvet beans, recorded a maximum of 25 percent mortality in chicks, which occurred in the first week when chicks were fed diets containing 50 percent velvet beans. Riley (1961) observed that chicks maintained on diets containing 90 percent raw field beans were very toxic. Mortality reached 80 to 90 percent by the ninth or tenth day. Our results are in agreement with those published by Harms et al. (1961) and Riley (1961).

An increase in unheated pinto bean content of the diet resulted in a greater depression of growth. This would indicate a greater content of growth depression in the diets containing the higher levels of pinto beans. This result is in agreement with the results obtained by Harms et al. (1961) while working on velvet beans.

Heating pinto beans with live steam improved the performance of chicks in all instances. An extreme imbalance of amino acid composition in the beans, or a marked deficiency of some nutrient may account for part of the growth depression, but improvement by heating with live steam suggests the presence of a growth inhibitor or toxic factors. These observations are in agreement with the observations of Kienholz et al. (1962) while working on legume seeds.

No marked difference was observed in feed efficiency with low levels of the heated pinto bean diet and the control diet.

In Experiment 2, the heat used in pelleting the ground beans or the complete formula did not appear to be sufficient to remove the growth depressant effect of the pinto beans, since they did not gain as well as those of the heated group of the same level. Heat treatment of beans with live steam at 100°C for 35 minutes was found to be beneficial to remove the toxic effect of the beans. Wagh et al. (1963), while working on red kidney beans, observed that 30 minutes of autoclaving at 121°C was sufficient to destroy all the antitryptic and hemagglutinating activity of the beans. Riley (1961), working on field beans, observed that dry autoclaving beans for 60 minutes at 10 p.s.i. or 30 minutes in the presence of 10 or 20 percent water optimumly improved growth.

When stabilized animal fat was added to diets containing either 10 or 20 percent heated pinto beans, a small but nonsignificant difference in weight gain was observed when compared with chicks fed the same diets without fat. The slight improvement in weight gain and feed efficiency observed by the addition of fat probably is due to: (1) increasing the efficiency of utilization of metabolizable energy; (2) improving the utilization of dietary protein; and/or (3) providing an unidentified growth factor. However, Riley (1961) observed that when the diet was supplemented with tallow, the growth rate decreased. He concluded that perhaps as the energy level was increased, a protein or essential amino acid deficiency developed, thus suggesting that the protein quality may be the limiting factor in the nutritive value of field beans.

Supplementation of raw pinto bean diets with DL-methionine, adjusting to the level of 0.8 percent, similar to the heated diets of the same level, did not give as much growth as that of heated ones with methionine supplementation. Woods (1952), while studying the effects of field beans as a dietary source of protein for rats, noted that raw beans could not be used for rats as a source of protein. He further observed that added DL-methionine improved growth obtained from cooked beans, but not from raw beans. Elvehjem (1958) observed that increasing the level at which an incomplete protein was fed, or supplementing it with single amino acids, might create such an imbalance with respect to other amino acids that the result might be less desirable than if the deficient protein were fed at low levels. Saxena et al. (1962) observed that supplementing the raw soybean meal diets with an amino acid mixture improved growth and feed efficiency.

The improvement, however, did not equal that obtained with the heated soybean meal diets supplemented with either DL-methionine or the amino acid mixture. The growth response obtained by an amino acid mixture suggested that amino acid availability was a factor in the growth depressing activity of raw soybean meal, but not the only factor.

It is concluded, therefore, that besides methionine there may be other limiting amino acids responsible for growth depression. If this hypothesis is true, one would expect a difference in amino acid composition between heated and unheated beans. The results obtained in the present investigation (Table 3) clearly indicates the amino acid content is more in heated than in unheated pinto beans. This observation also supports our findings in weight gain observed by feeding unheated and heated pinto beans.

SUMMARY AND CONCLUSIONS

Two experiments were conducted in order to observe the comparison of feeding heated and non heated pinto bean meal to broiler-strain chicks. A total of 548 straight-run Cobb's strain-cross White Rock day-old broiler-strain chicks were used in the two experiments. The chicks were kept in electrically heated battery brooders, with raised wire floors, to four weeks of age. At four weeks, they were transferred to unheated growing batteries. Body weight and feed consumption data were taken at two-week intervals during the eight-week period of the experiment.

The Kansas State University corn-sorghum grain-soybean oil meal basal broiler ration was used as the control diet. In Experiment 1, heated and unheated pinto beans were used at the following levels: 7.5, 15 and 52 percent. In Experiment 2, the level of pinto beans used was: 10, 15 and 20 percent. All diets used in Experiments 1 and 2 were isonitrogenous. In Experiment 2, the methionine and cystine level in all diets was adjusted to the standard of 0.8 percent by supplementing the required amino acid. Diets G and J were adjusted to the calorie level of the control diet by the addition of stabilized animal fat, while the calorie level in other diets differed one from another.

Results from these experiments indicated that the inclusion of unheated pinto beans in the diet at a level up to 20 percent did not produce growth retardation, and did not differ statistically when compared to the corn-sorghum grain-soybean oil meal control diet. However, it was noted that unheated pinto beans gave lesser growth. As the level of pinto beans increased in the diet, growth depression simultaneously increased. Heating the beans with live steam at 100°C for 35 minutes improved the weight gains. Heat required in processing feed for formulating rations was found inadequate to remove the deleterious effect of the beans. Higher levels of unheated pinto beans, such as 52 percent, were found to be toxic. The higher levels resulted in maximum growth depression when compared with other lots in the experiment, and caused 40 percent mortality. The mortality occurred in the first part of the second week. Addition of fat

to 10 or 20 percent heated pinto bean diets did not bring any significant difference in weight gain when compared with chicks fed the same diets without fat. However, diets containing fat were significantly better than the corn-sorghum grain-soybean oil meal control diet. Supplementing the raw pinto bean diets with DL-methionine to adjust the level of methionine and cystine to the required standard of 0.8 percent did not support growth equal to that of diets containing heated pinto beans at the same level. It suggests, therefore, that there appears to be some factor(s) in pinto beans which interfere(s) with normal processes of growth in chicks.

The following conclusions were drawn from the results of these experiments.

Addition of heated or unheated pinto beans in the chick ration depressed growth.

Growth depression was more severe in cases of unheated pinto beans.

Low levels of unheated pinto beans up to 20 percent were not toxic.

As the level of heated or unheated pinto beans was increased in the chick diet, growth depression gradually increased.

Very high levels of unheated pinto beans were toxic and caused 40 percent mortality which occurred in the early part of the second week.

Heat treatment improved the growth performance to some extent by inactivating the toxic substance in the raw bean that depressed growth. Heating the pinto beans with live steam at 100°C for 35 minutes was found to be beneficial.

Heat required in pelleting ground pinto beans and pelleting the complete formula was insufficient to remove the deleterious effect of the beans.

Addition of fat to heated pinto bean diets did not bring significant growth improvement over the diets containing the same levels of heated pinto beans without fat.

Supplementation of the raw pinto bean diet with DL-methionine did not support growth to the same extent as that of heated pinto beans at the same level with supplementation of DL-methionine.

There appears to be some factor(s) in pinto beans that interfere(s) with normal processes of growth in chicks.

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LITERATURE CITED

- Almquist, H. J. Unusual feedstuffs for poultry. Nulaid News, pp. 11-22. July, 1938.
- Amino acids and proteins. (D. M. Greenberg, ed.) p. 586. Charles C. Thomas, Springfield, Illinois, 1951.
- Almquist, H. J., and J. B. Merritt.

 Effect of soybean antitrypsin on experimental amino acid deficiency in the chick. Arch. Biochem. and Biophys. 31: 450-453, 1951.
 - Effect of soybean antitrypsin on growth of the chick. Arch. Biochem. and Biophys. 35:352-354, 1952a.
 - Effect of raw soybean meal on growth of the chicks. Proc. Soc. Expt. Biol. Med. 79:277-279, 1952b.
 - Effect of crystalline trypsin on the raw soybean growth inhibitor. Proc. Soc. Expt. Biol. Med. 83:269, 1953a.
- Accentuation of dietary amino acid deficiency by raw soybean growth inhibitor. Proc. Soc. Expt. Biol. Med. 84:333-334, 1955b.
- Almquist, H. J., E. Meechi, F. H. Kratzer, and C. R. Grau. Soybean protein as a source of amino acids for chicks. J. Nutrition 24:385-392. 1942.
- Altschul, A. M. Processed plant protein foodstuffs. Academic Press Inc., Publishers, New York, 1958.
- Alumot, E., and Zafrira Nitsan. The influence of soybean antitrypsin on the intestinal proteolysis of the chick. J. Nutrition 73:71-77, 1961.
- Armbruster, G., and H. C. Murray.

 The effect of canning procedures on the nutritive value of
 the protein in peas. J. Nutrition 44:205-216, 1951.
- Barnes, R. H., G. Fiala, and E. Kwong. Methionine supplementation of processed soybeans in rats. J. Nutrition 77:278-284, 1962.

- Bletner, J. K., N. E. Chalhaub, and O. E. Goff. Horse bean (Vicia faba L) as a vegetable protein in chick diets. Poultry Sci. 42:562-568, 1963.
- Block, R. G., and H. H. Mitchell.

 The correlation of the amino acid composition of proteins with their nutritive value. Nutrition Abstr. and Rev. 16: 249-278. 1946.
- Booth, A. N., D. J. Robbins, Wm. E. Ribelin, and F. De Eds. Effect of raw soybean meal and amino acids on pancreatic hypertrophy. Proc. Soc. Expt. Biol. Med. 104:681-683, 1960.
- Borchers, R. Concerning the site of nitrogen absorption in rats fed autoclaved or raw soybean oil meal. Science 117:482, 1953.
- Counteraction of the growth depression of raw soybean oil meal by amino acid supplements in weaning rats. J. Nutrition 75:350-354, 1961.
- Borchers, R., and C. W. Ackerson.
 The nutritive value of legume seeds. J. Nutrition 41:339-345, 1950.
- Borchers, R., C. W. Ackerson, and R. M. Sandstedt.
 Trypsin inhibitor III. Determination and heat destruction of the trypsin inhibitor of soybeans. Arch. Biochem. 12: 367-374, 1947.
- Borchers, R., D. Mohammad-Abadi, and J. M. Weaver. Antibiotic growth stimulation of rats fed raw soybean oil meal. Agri. and Feed Chem. 5:371-373, 1957.
- Borchers, R., C. W. Ackerson, F. E. Mussehl, and A. Moehl. Trypsin inhibitor VIII. Growth inhibiting properties of a soybean trypsin inhibitor. Arch. Biochem. 19:317-322, 1948.
- Bowman, D. E.
 Fractions derived from soybeans and navy beans which retarded tryptic digestion of casein. Proc. Soc. Expt. Biol. Med. 57: 159-140, 1944.
 - Differentiation of soybean antitryptic factor. Proc. Soc. Expt. Biol. Med. 63:547-550, 1946.
 - Further differentiation of bean trypsin inhibiting factors. Arch. Biochem. 16:109-113, 1948.
 - Characteristics of proteose trypsin inhibitor of soybeans. Federation Proc. 9:155, 1950.

- Carrol, R. W., G. W. Hensley, and W. R. Graham. The site of nitrogen absorption in rats fed raw and heat treated soybean meals. Science 115:36-39, 1952.
- Chernick, S. S., S. Lepkovsky, and I. L. Chaikoff.

 A dietary factor regulating the enzyme content of the pancreas, changes induced in size and proteolytic activity of the chick pancreas by the ingestion of raw soybean meal.

 Am. J. Physiol. 155:33, 1948.
- Clandinin, D. R., W. W. Cravens, J. G. Halpin, and E. B. Hart. Supplementary value of methionine, cystine and choline in a practical soybean oil meal starter ration. Poultry Sci. 25:509-516. 1946.
- Elvehjem, C. A. Amino acid balance in nutrition. J. Am. Dietet. Assoc. 32:305-308, 1956.
- Evans, R. J., and J. McGinnis. The influence of autoclaving soybean oil meal on the availability of cystine and methionine. J. Nutrition 31:449-461, 1946.
 - Cystine and methionine metabolism by chicks receiving raw or autoclaved soybean oil meal. J. Nutrition 35:477-487, 1948.
- Evans, R. J., A. C. Grochke, and H. A. Butts. Studies on the heat inactivation of cystine in soybean oil meal. Arch. Biochem. 30:414-422, 1951.
- Everson, G. J., and A. Heckert. The biological value of some leguminous sources of protein. J. Am. Dietet. Assoc. 20:81-82, 1944.
- Finks, A. J., and C. O. Johns. The nutritive value of the proteins from the Chinese and Georgia velvet beans. Am. J. Physiol. 57:61-67, 1921.
- Fisher, H., and D. Johnson, Jr.

 The effectiveness of essential amino acid supplementation in overcoming the growth depression of unheated soybean meal.

 Arch. Bioch. and Biophys. 77:124-128, 1958.
- Fisher, H., D. Johnson, Jr., and S. Ferdo. The utilization of raw soybean meal protein for egg production in chickens. J. Nutrition 61:611-621, 1957.
- Freeman, G. F.
 Southwestern beans and taperies. Arizonia Agri. Expt. Sta.
 Bul. 68, 1912.

- Gracia, Fabian.

 New Mexico beans. New Mexico Agri. Expt. Sta. Bul. 105, 1917.
- Greenwood, Mary L.
 Pinto beans: Their preparation and palatability. New Mexico Agri. Exrt. Sta. Bul. No. 231, 1935.
- Ham, W. E., and R. M. Sandstedt. A proteolytic inhibiting substance in the extract from unheated soybean meal. J. Biol. Chem. 154:505-506, 1944.
 - Ham, W. E., R. M. Sandstedt, and F. E. Mussehl. The proteolytic inhibiting substance in the extract from unheated soybean meal and its effect upon growth in chicks. J. Biol. Chem. 161:635-642, 1945.
 - Harms, R. H., Charles F. Simpson, and P. W. Waldroup. Influence of feeding various levels of velvet beans to chicks and laying hens. J. Nutrition 75:127-131, 1961.
 - Hayward, J. W., and F. H. Hafner.

 The supplementary effect of cystine and methionine upon the protein of raw and cooked soybeans as determined with chicks and rats. Poultry Sci. 20:139-150, 1941.
 - Hayward, J. W., H. Steenbock, and G. Bohstedt. The effect of heat as used in the extraction of soybean oil upon the nutritive value of the protein of soybean oil meal. J. Nutrition 11:219-234, 1936.
 - The effect of cystine and casein supplements upon the nutritive value of the protein of raw and heated soybeans.
 J. Nutrition 12:275-283, 1936.
 - Honavar, P. M., Cheng-Ven Shih, and I. E. Liener. Inhibition of the growth of rats by purified hemagglutinin fractions isolated from Phaseolus vulgaris. J. Nutrition 77: 109-114, 1962.
 - Inamdar, and Sohoni.
 Ann. Biochem. and Exptl. Med. 21:199, 1961. Original reference not found. Abstract taken from growth inhibitors in soybeans—A review. Feedstuff Vol. 35(17):44-46, April 20, 1963.
 - Ingram, G. R., W. H. Riesen, W. W. Cravens, and C. A. Elvehjem. Evaluating soybean oil meal protein for chick growth by enzymatic release of amino acids. Poultry Sci. 28:898-902, 1949.
 - Jaffe, W. G. Limiting essential amino acids in some legume seeds. Proc. Soc. Expt. Biol. Med. 71:398-399, 1949a.

- Jaffe, W. G. Toxicity of raw kidney beans. (In German, English summary.) Experientia 5.81. 1949. (Original not available for examination: abstracted in Nutri. Abst., Rev. 19:1307, 1949b.)
 - Protein digestibility and trypsin inhibitor activity of legume seeds. Proc. Soc. Expt. Biol. Med. 75:219-220, 1950.
 - Inhibition of growth of rats caused by some legume seeds. (In German, English summary.) Actacient. venzolana 1,62-64, 1950. (Original not available for examination: abstracted in Nutri. Abst., Rev. 20:5601, 1951a.)
- _____.Toxicity of raw beans for rabbits. (In German,
 English summary.) Actacient, venzolana 1,16-17, 1950.
 (Original not available for examination: abstracted in Nutri.
 Abst. Rev. 20:6140, 1951b.)
- Jaffe, W. G., P. Plachart, J. L. Pacz Pumar, R. Torrelba, and D. N. Franceschi.
 A toxic substance in raw beans (Phaseolus vulgaris). (In German, English summary.) Arch. Venezol Nutricon, 1955, 6, 195-205. (Original not available for examination: abstracted in Nutri. Abst. Rev. 27:3962, 1957.)
- Jensen, Leo S., and H. C. Saxena. Growth inhibitors in soybean--A review. Feedstuffs, Vol. 35 (17):44-46, 1963.
- Johns, C. O., and A. J. Finks.

 The role of cystine in nutrition as exemplified by nutrition experiments with the proteins of navy bean, Phaseolus vulgaris. J. Biol. Chem. 41:379-389, 1920.
- Johnson, L. M., H. T. Parsons, and H. Steenbock. The effect of heat and solvents on the nutritive value of soybean protein. J. Nutrition 18:423-434, 1939.
- Kakde, M. L., and R. J. Evans. Growth inhibition of rats fed raw navy beans. Federation proceedings, Vol. 22, No. 2, Part 1. Abst. 2333, March-April, 1963.
- Kienholz, E. W., Leo S. Jensen, and J. McGinnis. Evidence for chick growth inhibitors in several legume seeds. Poultry Sci. 41:367-371, 1962.
- Klose, A. A., J. D. Greaves, and H. L. Fevold. Inadequacy of proteclytic enzyme inhibition as explanation for growth depression by lima bean protein fractions. Science 108:88-69, 1948.

- Klose, A. A., B. Hill, and H. L. Fevold. Presence of a growth inhibiting substance in raw soybeans. Proc. Soc. Expt. Biol. Med. 62:10-12, 1946.
- Klose, A. A., B. Hill, J. D. Greaves, and H. L. Fevold. Growth depressing fractions in raw lima beans. Arch. Biochem. 22:125-223. 1949.
- Kuiken, K. A., and C. M. Lyman. Essential amino acid composition of soybean meals prepared from twenty strains of soybeans. J. Biol. Chem. 177:29-36, 1949.
- Kunitz, M. Crystallization of a trypsin inhibitor from soybeans. Science 101:668-669, 1945.
- Crystalline soybean trypsin inhibitor. J. Gen. Physiol. 29:149-154. 1946.
- Crystalline soybean trypsin inhibitor. J. Gen. Physiol. 30:291-310, 1947a.
- Isolation of crystalline protein compound of trypsin and soybean trypsin inhibitor. J. Gen. Physiol. 30:311-320, 1947b.
- The kinetics and thermodynamics of reversible denaturation of crystalline soybean trypsin inhibitor. J. Gen. Physiol. 32:241-269, 1948.
- Lantz, E. M., H. W. Gough, and A. M. Campbell. Effects of planting date on the composition and cooking quality of pinto beans. New Mexico Agri. Expt. Sta. Bul. No. 467, 1962.
- Lepkovsky, S., E. Bingham, and R. Pencharz.

 The fate of proteolytic enzymes from the pancreatic juice of chicks fed raw and heated soybeans. Poultry Sci. 38:1289-1295, 1959.
- Liener, I. E. Soyin a toxic protein from the soybean. J. Nutrition 49: 527-539, 1953.
- Liener, I. E., H. J. Denuel, Jr., and H. L. Fevold.

 The effect of supplemental methionine on the nutritive value of the diets containing concentrates of the soybean trypsin inhibitor. J. Nutrition 39:325-339, 1949.

- Liener, I. E., and S. Wada. Liver-xanthine oxidase activity in relation to availability of methionine from soybean protein. Proc. Soc. Expt. Biol. Med. 82:484-486, 1955.
- Lenerode, P. A., P. E. Waibel, and B. S. Pomery. Effects of antibiotics, amino acids, zinc and enzymes on the growth depression in turkey produced by raw scybeans. J. Nutrition 75:427-434, 1961.
- McGinnis, J., and R. J. Evans. Amino acid deficiencies of raw and over heated soybean oil meal for chicks. J. Nutrition 34:725-732, 1947.
- Melnick, D., B. L. Oser, and S. Weiss. Rate of enzymic digestion of proteins as a factor in nutrition. Science 103:326-329, 1946.
- Mitchell, H. H., T. S. Hamilton, and J. R. Beadler.

 The importance of commercial processing for the protein value of food products. J. Nutrition 29:13-25, 1945.
- Mohon, J. H., and R. H. Common.

 The amino acid composition of the horse bean (Vicia faba L)
 as related to its nutritive value for chicks. Sci. Agric.
 30:43-49, 1950.
- Murray, H. C.

 The biological value of the protein of field pea products with a comparison of several methods used for this determination. J. Nutrition 35:257-267, 1948.
- Nesheim, M. C., J. D. Garlich, and D. T. Hopkins. Studies on the effect of raw soybean meal on fat absorption in young chicks. J. Nutrition 73:89-94, 1962.
- Osborne, T. B., and L. B. Mendel.

 The use of soybean as food. J. Biol. Chem. 32:369-387, 1917.
- Oser, B. L.

 Method for integrating essential amino acid content in the nutritional evaluation of protein. J. Am. Dietet. Assoc. 27:396-402, 1951.
- Read, J. W. Nutritional value of the Georgia velvet bean (Stizolobium Deeringnum). J. Agri. Res. 24:433, 1923.
- Richardson, L. R.
 Southern peas and other legume seeds as a source of protein for the growth of rats. J. Nutrition 36:451-462, 1948.

- Riesen, W. H., D. R. Clandinin, C. A. Elvehjem, and W. W. Cravens. Liberation of essential amino acids from raw, properly heated, and over heated soybean oil meal. J. Biol. Chem. 167:143-150, 1947.
- Riley, D. J.
 Studies on the nutritional value of some little used protein feedstuffs for poultry. Unpublished M.S. Thesis, Washington State University Library, 1961.
- Russell, W. C., M. W. Taylor, T. G. Mehrhof, and R. R. Hirch.
 The nutritive value of the protein of varieties of legumes
 and the effect of methionine supplementation. J. Nutrition
 32:313-326, 1946.
- Salmon, W. D.
 The effect of feeding velvet beans to pigeons. Science 56: 141, 1922.
- Saxena, H. C., Leo S. Jensen, and J. McGinnis. Influence of dietary protein level on chick growth depression by raw soybean meal. J. Nutrition 77:241-244, 1962a.
 - Failure of amino acid supplementation to completely overcome the growth depression effect of raw soybean meal in chicks. J. Nutrition 77:259-263, 1962b.
 - Influence of raw soybeans on oxygen consumption and liver and muscle glycogen content of chicks. Poultry Sci. 41:1304-1305. 1962c.
- Pancreatic hypertrophy and chick growth inhibition by soybean fractions devoid of trypsin inhibitor. Proc. Soc. Exptl. Biol. Med. 112:101-105, 1963.
- Saxena, H. C., Leo S. Jensen, J. McGinnis, and J. K. Lauber. Histophysiological studies of chick pancreas as influenced by feeding raw soybean meal. Proc. Soc. Expt. Biol. Med. 112: 390-393, 1963.
- Schneider, B. H., and D. F. Miller.
 Biological value of pea protein. J. Nutrition 52:581-590, 1954.
- Scott, J. M. Pork production in Florida. Florida Agri. Expt. Sta. Bul. No. 141, 1917.

- Sherwood, F. W., V. Veldon, and W. J. Peterson. Effect of cooking and of methionine supplementation on the growth promoting properties of cow pea (Vigua Simensis). J. Nutrition 52:199-208, 1954.
- Simmons, W. M.

 Vegetable section Economics and Statistical Analysis Division U.S.D.A., Washington, D.C. By personal communication. May 2, 1963.
- Snedecor, G. W. Statistical methods. The Iowa State University Press, Ames, Iowa, Fifth edition, 1956.
- Sohoni, K., U. Apte, and K. S. Ambe. Trypsin inhibitors in Indian foodstuffs. Effect of raw double bean (Faba vulgaris M) and double bean trypsin inhibitor on the growth of rats. J. Sci. Ind. Res. 17(c):42-46, 1958.
- Sure, B., and J. W. Read. Biological analysis of the seed of the Georgia velvet bean, Stizolobium-Deeringianum. J. Agri. Res. 22:5, 1921.
- Tauber, H., B. B. Kershaw, and R. D. Wright. Studies on the growth inhibitor fraction of lima beans and isolation of a crystalline heat stable trypsin inhibitor. J. Biol. Chem. 179:1155-1161, 1949.
- Tempelton, G. S.

 Improving the quality of peanut fed hogs by finishing in dry lot on corn and tankage, corn and cottonseed meal, corn and velvet beans. Alabama Expt. Sta. Bul. No. 213, 1920.
- Tempelton, G. S., H. C. Ferguson, and E. Gibbons.

 Velvet beans compared with cotton seed meal for fattening steers, Part I. Velvet beans, cottonseed meal and corn as feed for dairy cattle, Part II. Velvet bean pasture compared with corn and dried blood, velvet bean meal compared with corn for fattening hogs, Part III. Alabama Expt. Sta., Auburn, Ala. Bul. 198, 1917.
- Titus, H. W.

 Nutritive properties of pinto beans and pinto bean straw and their use as feed for cattle. New Mexico Agri. Expt. Sta. Bul. No. 143, 1924.

The scientific feeding of chickens. The Interstate printers and publishers Inc., Danville, Illinois. Fourth edition, 1961.

- Wagh, P. V., D. F. Klaustermeier, P. E. Waibel, and I. E. Liener. Nutritive value of red kidney beans. J. Nutrition 80:191-195, 1963.
- Waterman, H. C., and C. O. Johns. Studies on the digestibility of protein in vitro. The effect of cooking on the digestibility of phaseolin. J. Biol. Chem. 46:9-17. 1921.
- Waterman, H. C., and D. B. Jones. Studies on the digestibility of proteins in vitro. The relative digestibility of various preparations of the proteins from the Chinese and Georgia velvet beans. J. Biol. Chem. 47:285-295. 1921.
- Westfall, R. J., and S. M. Hauge.

 The nutritive quality and the trypsin inhibitor content of soybean flour heated at various temperatures. J. Nutrition 35:379-389, 1948.
- Wilgus, H. S., Jr., L. C. Norris, and G. F. Heuser. Effect of heat on nutritive value of soybean oil meal. Ind. Eng. Chem. 28:586-588, 1936.
- Wilson, C. P., and J. L. Lantow. Bean feeding. New Mexico Agr. Expt. Sta. Bul. 155, 1926.
- Woods, E. Field beans as a dietary source of protein. University of Idaho Agri. Expt. Sta. Bul. No. 289, 1952.
- Woods, E., W. M. Beeson, and D. W. Bolin. Field peas as a source of protein for growth. J. Nutrition 26:327-335. 1943.



Table 1. Composition of diets (Experiment 1).

| | : | Ra | tion | |
|---|-------|-------|-------|-------|
| | : A | : B | ; C | : D |
| | : | | % | |
| Corn, yellow, ground | 30.0 | 30.0 | 30.0 | 30.0 |
| Sorghum grain, ground | 31.4 | 26.8 | 22.4 | 0.0 |
| Soybean oil meal, solv. ext. | 28.6 | 25.7 | 22.6 | |
| Pinto beans | 0.0 | 7.5 | 15.0 | |
| Alfalfa meal, 17% prot., dehyd. | 2.0 | 2.0 | 2.0 | 2.0 |
| Fish meal, menhaden | 4.0 | | | |
| Midsol(R) | 1.5 | | | |
| Calcium carbonate | 1.0 | 1.0 | | |
| Dicalcium phosphate | 1.0 | 1.0 | 1.0 | |
| Sodium chloride (salt) | 0.5 | 0.5 | 0.5 | 0.5 |
| Total | 100.0 | 100.0 | 100.0 | 100.0 |
| | | | | |
| Added per 100 pounds of ration | | - | gm | |
| Trace mineral mix2 | 23 | 23 | 23 | 23 |
| Vit. A (10,000 U.S. units/gm) | 10 | 10 | 10 | 10 |
| Vit. D3 (15,000 I.C.U./gm) | 5 | 5 | 5 | 5 |
| B-complex vit. mix3 | 46 | 46 | 46 | 46 |
| D-L methionine (Hydan (R)) | 23 | 23 | 23 | 23 |
| Choline chloride, 25% mix Baciferm - 10(R) | 40 | 40 | 40 | 40 |
| Baciferm - 10(R) | 11.5 | 11.5 | 11.5 | 11.5 |
| Tylan - $10(R)$ | 11.5 | 11.5 | 11.5 | 11.5 |
| Pro-Gen 90(R) | 23 | 23 | 23 | 23 |
| nf-180(R) | 23 | 23 | 23 | 23 |

By calculation, diet A contained 21% protein. Diets B, C, and D were isonitrogenous.

 $^{^2}$ Trace mineral premix supplying by percent Mn 10; Fe 10; Ca, max 14, min 12; Cu 1; Zn 5; $\rm I_2$ 0.3; Co 0.1.

B-complex vitamin mix supplying in mg/lb: riboflavin 20,000; pantothenic acid 2,680; niacin 6,000; choline chloride 20,000.

⁽R) Registered trade-mark.

Table 2. Composition of diets1 (Experiment 2).

| | | | Rat | Lon | | |
|--|-------|-------|-------|-------|-------|-------|
| : | E : | F | | | I | J |
| | | | 9 | 6 | | |
| Corn, yellow, ground | 30.0 | 30.0 | 30.0 | 30.0 | 30.0 | 30.0 |
| Sorghum grain, ground Soybean oil meal, solv. | 31.4 | 25.3 | 21.6 | 22.4 | 19.3 | 12.6 |
| ext. | 28.6 | 24.7 | 25.4 | 22.6 | 20.7 | 22.0 |
| Pinto beans Alfalfa meal, 17% prot., | 0.0 | 10.0 | 10.0 | 15.0 | 20.0 | 20.0 |
| dehyd. | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 |
| Fish meal, menhaden | 4.0 | 4.0 | 4.0 | 4.0 | 4.0 | 4.0 |
| Midsol(R) | 1.5 | 1.5 | 1.5 | 1.5 | 1.5 | 1.5 |
| Calcium carbonate | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 |
| Dicalcium phosphate | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 |
| Sodium chloride (salt) | 0.5 | 0.5 | 0.5 | 0.5 | 0.5 | 0.5 |
| Fat | 0.0 | 0.0 | 3.0 | 0.0 | 0.0 | 5.4 |
| Total | 100.0 | 100.0 | 100.0 | 100.0 | 100.0 | 100.0 |
| Added per 100 pounds of I | ation | | _ | gm | | |
| Trace mineral mix2 | 23 | 23 | 23 | 23 | 23 | 23 |
| Vit. A (10,000 U.S.P. units/gm) | 10 | 10 | 10 | 10 | 10 | 10 |
| Vit. D3 (15,000 I.C.U./ | 5 | 5 | 5 | 5 | 5 | 5 |
| gm) B-complex vit. mix ³ | 46 | 46 | 46 | 46 | 46 | 46 |
| D-L methionine (Hydan(R)) | | 23.08 | 23.07 | 23.10 | 23.11 | |
| Choline chloride. | 20.04 | 20.00 | 20.01 | 20.10 | 20.11 | 20. |
| 25% wir | 40 | 40 | 40 | 40 | 40 | 40 |
| Baciferm - 10(R) | 11.5 | 11.5 | 11.5 | 11.5 | 11.5 | 11. |
| Tylan - 10(R) | 11.5 | 11.5 | 11.5 | 11.5 | 11.5 | 11. |
| Pro-Gen 90(R) | 23 | 23 | 23 | 23 | 23 | 23 |
| Pro-Gen 90(R) nf-180(R) | 23 | 23 | 23 | 23 | 23 | 23 |
| | | | | | | |

 $^{^{\}rm l}$ By calculation, diet E contained 21% protein. Diets F, G, H, I and J were isonitrogenous.

² Trace mineral premix supplying by percent Mn 10; Fe 10; Ca, max. 14, min. 12; Cu 1; Zn 5; I₂ 0.3; Co 0.1.

³ B-complex vitamin mix supplying in mg/lb: riboflavin 2,000; pantothenic acid 2,680; niacin 6,000; choline chloride 20,000.

⁽R) Registered trade-mark.

Table 3. Essential amino acid composition of pinto beans and calculated analysis of essential amino acids in the control, 15 and 52 percent pinto bean diets used in Experiment 1.

| Amino acids | Pinto beans | Pinto beans heated | : oil : | Pinto beans eated 15% | Pinto beans heated 52% | Pinto beans un- heated 52% |
|--|---|--|--|--|---|---|
| Arginine Histidine Isoleucine Leucine Lysine Methionine Half cystine Phenylalanine Tyrosine Threonine Valine Glycine | 1.030 0.471 0.742 1.271 1.151 0.155 0.062 0.858 0.360 0.614 0.777 | 1.248 0.554 0.915 1.639 1.340 0.209 0.157 1.188 0.482 0.799 1.096 0.840 | 0.570 0.991 2.101 1.259 0.313 0.297 1.165 0.733 0.882 2.404 | 1.240 0.500 0.909 1.846 1.170 0.199 0.297 0.932 0.559 0.812 1.101 1.086 | 1.264 0.543 0.878 1.726 1.278 0.285 0.201 1.145 0.613 0.815 1.078 | 1.304 0.585 0.884 1.728 1.340 0.252 0.114 1.091 0.627 0.808 1.096 |

 $[\]ensuremath{\mathbf{l}}$ Result of single analysis. Percent amino acids on the basis of the sample with no moisture correction.

Table 4. Analysis of raw pinto beans, edible part per 100 gm.

| Protein | 23.0 | gm |
|--------------------|---------|----|
| Fat | 1.2 | |
| Total carbohydrate | 63.7 | 11 |
| Fiber | 3.9 | 11 |
| Ash | 4.0 | 11 |
| Moisture | 8.1 | 17 |
| Calcium | 163.0 | mg |
| Phosphorus | 437.0 | " |
| Iron | 6.9 | 11 |
| Thiamin | 0.65 | 11 |
| Riboflavin | 0.24 | 11 |
| Niacin | 2.0 | 11 |
| Ascorbic acid | 2.0 | 11 |
| Calories | 349/100 | gm |

Table 5. Analysis of variance for chick weight (Experiment 1).

| | 2 | | | |
|----------------------|----------------------------------|------------|----------------|----------------|
| Source of variation | :Degrees: : of : :freedom: | Sum : | Mean square | : : F-ratio |
| Diet | 6 | 794192.76 | 132365.46 | 5.62** |
| Sex | 1 | 727828.20 | 727828.20 | 30.92** |
| Diet sex interaction | 6 | 1769959.20 | 294993.20 | 12.53** |
| Within sub class | 158 | 3718462.91 | 23534.57 | |
| Total | 171 | 7010443.07 | | |

Ranked lots by Fisher's L.S.D. method for chick weight
L.S.D. 113 for 5% level of probability

D D₁ C B C₁ B₁ A
Treatment means 827 1160 1220 1271 1283 1291 1340

^{**} Significant at 1% level of probability.

 $^{^{\}rm l}$ Any two lots not underscored by the same line are significantly different at 5% level of probability.

Analysis of variance for chick weight (Experiment 2). Table 6.

| Source of variation | riation | • •• | of freedo | of freedom | : Sum | Sum of squares | ares | ı | Mean square | | F-ratio | tio |
|----------------------|--|----------------|-----------|------------|---------------------------------------|----------------|--------|-------|-------------|------|---------|----------|
| Diet | | | | 11 | Ō | 978461.11 | 11 | ω | 88251.01 | d | 7. | 7.52** |
| Sex | | | | ٦ | 52 | 5297699.23 | 23 | 586 | 5297699.23 | ю | 448. | 448.31** |
| Diet sex interaction | tion | | | 11 | 4 | 449239.22 | 22 | 4 | 40839.93 | ю | 8. | 3,45** |
| Within sub class | | | 63 | 341 | 40 | 4029593.78 | 78 | П | 11816.99 | 6 | | |
| Total | | | 6.9 | 364 | 107 | 10754993.34 | 34 | | | | | |
| | Ranked lots by Fisher's L.S.D. method for chick weight | ots] | by Fis | her's | L.S.D. | metho | d for | ch1ck | weight | | | |
| | | L.S.J | 0. 77 | for 5% | L.S.D. 77 for 5% level of probability | of pr | obabil | lty | | | | |
| | н | H ₃ | н | Ι | Īω | $^{\rm H}_2$ | 戶 | I_1 | H4 | FJ | Ö | ٦ |
| Treatment means | 1088 | 1100 | 1115 | 1121 | 1148 | 1159 | 1159 | 1193 | 1201 | 1225 | 1236 | 1269 |
| | • | | | | | | | | | | | |
| | | | | | | | | | | | | |
| | | | | | | | | | | | | |

** Significant at 1% level of probability.

Any two lots not underscored by the same line are significantly different at 5% level of probability.

Table 7. Analysis of feed conversion (Experiment 1).

| Source of variation | : | Degrees of freedom | : : : of | Sum squares | : | Mean square | : | F-ratiol |
|---------------------|---|--------------------------|----------------|----------------|---|----------------|---|-----------|
| Diets | | 4 | | 0.1860 | | .045 | | 3.75 n.s. |
| Within diets | | 5 | | 0.058 | | .012 | | |
| Total | | 9 | | 0.2440 | | | | |

Ranked lots 2 based on Fisher's L.S.D. method of analysis of feed conversions

A B₁ B C C₁
2.21 2.21 2.48 2.49 2.49

n.s. Nonsignificant at 5% level of probability.

L.S.D. 0.20 for 10% level of probability.

² Any two lots not underscored by the same line are significantly different at 10% level of probability.

Table 8. Analysis of feed conversion (Experiment 2).

| Source of variation | : Degrees : of freedom: | | Mean square | : | F-ratio |
|---------------------|-------------------------|--------|----------------|---|-----------|
| Diets | 11 | 0.1106 | 0.0101 | | 1.55 n.s. |
| Within diets | 12 | 0.0780 | 0.0065 | | |
| Total | 23 | 0.1886 | | | |

n.s. Nonsignificant at 5% level of probability.

Effects of heated and unheated pinto beans on chick weight gain (Experiment 1). Table 9.

| | ••• | | Male : weight : | Female weight | : weight | |
|----------------|---------------------------|------------------|-----------------|------------------|----------|--------------------|
| Ration | : Supplement to ration | Form of ration : | gain | gain | : gain | : Feed :efficiency |
| A | Soybean oil meal control | Mash | 1487 | 1193 | 1340 | 2.21 |
| В | 7.5% pinto beans unheated | Mash | 1387 | 1155 | 1271 | 2.48 |
| O | 15% pinto beans unheated | Mash | 1320 | 1120 | 1220 | 2,49 |
| В | 7.5% pinto beans heated | Mash | 1391 | 1191 | 1831 | 2.21 |
| c ₁ | 15% pinto beans heated | Mash | 1362 | 1205 | 1283 | 8.49 |
| D | 52% pinto beans unheated | Mash | 984 | 670 | 827 | 3.65 |
| D_1 | 52% pinto beans heated | Mash | 1298 | 1022 | 1160 | 3.18 |

Table 10. Effects of heated and unheated pinto beans on chick weight gain (Experiment 2).

| Ration | Supplement to ration | Form of ration | weight gain | . Female . weight . gain gm | : Average : weight : gain | Feed: Feed: |
|------------------------------|---|----------------|----------------|--------------------------------------|---------------------------------|-------------|
| н | 15% pinto beans unheated | Mash | 1235 | 995 | 3111 | 2.50 |
| $_{\rm H_1}$ | 15% pinto beans, beans pelleted reground | Crumble | 1227 | 949 | 1088 | 2.57 |
| Н2 | 15% pinto beans unheated, formula pelleted | Grumble | 1284 | 1034 | 1159 | 2.38 |
| Н3 | 15% pinto beans (beans pelleted and reground), formula pelleted | Crumble | 1194 | 1006 | 1100 | 8.46 |
| Н4 | 15% pinto beans heated live steam | Mash | 1346 | 1056 | 1201 | 2.44 |
| 国 | Soybean oil meal control | Mash | 1265 | 1053 | 1159 | 2.42 |
| ĒΨ | 10% pinto beans unheated, no fat | Mash | 1251 | 1045 | 1148 | 2.51 |
| Fl | 10% pinto beans heated, no fat | Mash | 1362 | 1088 | 1225 | 2.45 |
| 6 | 10% pinto beans heated, with fat | Mash | 1358 | 1114 | 1236 | 2.46 |
| н | 20% pinto beans unheated, no fat | Mash | 1241 | 1001 | 1121 | 29.2 |
| $^{\mathrm{I}}_{\mathrm{J}}$ | 20% pinto beans heated, no fat | Mash | 1330 | 1056 | 1193 | 2.52 |
| ы | 20% pinto beans heated, with fat | Mash | 1429 | 1109 | 1269 | 2.39 |

A COMPARISON OF FEEDING HEATED AND NON HEATED PINTO BEAN MEAL TO BROILER-STRAIN CHICKS

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AN ABSTRACT OF A MASTER'S THESIS

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MASTER OF SCIENCE

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Two experiments were conducted in order to observe the comparison of feeding heated and non heated pinto bean meal to broiler strain chicks. A total of 548 straight-run Cobb's strain-cross White Rock day-old broiler strain chicks were used in the two experiments. They were reared in batteries under normal poultry husbandry practices. The Kansas State University corn-sorghum grain-soybean oil meal basal broiler ration was used as the control diet. In Experiment 1, all feed was mixed at the university poultry farm, and in Experiment 2, the experimental diets were prepared at the university feed mill. The level of pinto beans used in the experimental diets was 7.5, 10, 15, 20 and 52 percent. All diets used in Experiments 1 and 2 were were isonitrogenous. In Experiment 2, the levels of methionine and cystine in the diet were adjusted to 0.8 percent by supplementation. Two diets containing 10 and 20 percent heated pinto beans were brought to the calorie level of the control by the addition of stabilized animal fat. The calorie level in other diets differed one from another.

Body weight and feed consumption data were taken at two-week intervals. The experiments were conducted for a period of eight weeks each.

From the analysis of data, the following conclusions were drawn:

- (1) Addition of heated or unheated pinto beans in the chick ration depressed growth.
- (2) Growth depression was more severe in cases of unheated pinto beans.

- (3) Low levels of unheated pinto beans up to 20 percent were not toxic.
- (4) As the level of heated or unheated pinto beans was increased in the chick diet, growth depression gradually increased.
- (5) Very high levels of unheated pinto beans were toxic and caused 40 percent mortality which occurred in the early part of the second week.
- (6) Heat treatment improved the growth performance to some extent by inactivating the toxic substance in the raw bean that depressed growth.
- (7) Heating the pinto beans with live steam at 100° C for 35 minutes was found to be beneficial.
- (8) Heat required in pelleting ground pinto beans or complete formula pelleting was insufficient to remove the deleterious effect of the beans.
- (9) Addition of fat to heated pinto bean diets did not bring significant growth improvement over the diets containing same levels of heated pinto beans without fat.
- (10) Supplementation of the raw pinto bean diet with DL-methionine did not support growth to the same extent as that of heated pinto beans at the same level with supplementation of DL-methionine.
- (11) There appears to be some factor(s) in pinto beans that interfere(s) with normal processes of growth in chicks.