EFFECT OF THE MAILLARD REACTION ON CHICK GROWTH AT FIVE LEVELS OF BROWNING AND A COMPARISON OF SOURCES OF FLOUR AND GLUCOSE IN UNBROWNED FEED

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CHAPTER 1

INTRODUCTION

Proteins are necessary to animals for tissue synthesis and regulation of many body functions. The quality and nutritive value of proteins are determined by the content and availability of essential amino acids (Robinson and Lawler, 1977).

Heat-induced browning of food has been shown to cause physicochemical and nutritive changes in the proteins of the food (Mauron, 1981). This browning called non-enzymatic browning or Maillard reaction, is important to food science because of its effect on flavor, color, and nutritive value of the food products (Baldwin, Lowry, and Thiessen, Jr., 1951). Nutritional concerns are because, during browning, carbohydrates (as reducing sugars) and amino acids, such as lysine, are bound and become unavailable to the ingesting animal (Lang, 1970; Adrian, 1974; Sgarbieri, Amaya, Tanaka, and Chichester, 1973a).

Several researchers have fed rats unheated and heated foods. They found that heat-damaged proteins affect growth in that animals fed severely heated food gained less (Frazier, Cannon, and Hughes, 1953; Kimiagar, Lee, and Chichester, 1980).

Van Soest has stated that the non-enzymatic browning reaction between amino acids and the degradation products of sugar in foods during heating (baking, frying, etc.) will increase the apparent yield of lignin, a component of dietary fiber. This "artifact" lignin, resulting from non-enzymatic browning, is recovered from neutral-detergent fiber or acid-detergent fiber residues when analyzing for dietary fiber. Breakfast cereals such as Wheat Chex, corn flakes, and other similar browned food products contain significant amounts of this "artifact" lignin which might have properties of dietary fiber such as water-holding capacity, ion-exchange capacity, bulk density, and bacterial fermentability which are important in nutrition (Van Soest, 1965 and 1978; Eastwood and Mitchell, 1976).

Knight and Hanson (1982, 1983) at Oklahoma State University conducted two trials attempting to determine whether there was significant binding of nutrients when chicks were fed a browned ration, based on whether degree of browning affected growth of chicks. In Trial 1, a chick feed was prepared using J. T. Baker Company U.S.P. dextrose, Pillsbury bread flour, egg solids, yeast, and oil. Portions of the feed were browned for 0, 20, and 30 minutes at 350^{o}F (176.7^{o}C), and both the 20 and 30 minute browned feed resulted in little or no chick growth.

In Trial 2, the feeding was repeated, but Cerelose brand glucose from the Corn Products Company and Multifoods

brand flour were used for the sugar and flour in preparing the feeds. The feeds were browned at 350°F (176.7°C) for 0, 5, 10, 15, and 20 minutes. From the results of Trial 1 and Trial 2, it was determined that browning did have an effect on growth, but the impact on growth was not as great, even at the 20 minute level, when the feed was prepared using the different sugar and flour combinations.

Although in both these trials, browning of the feeds appeared to decrease the utilization of nutrients and thus affect chick growth, additional information is needed to determine whether or not the browned products have the characteristics of fiber, the level of browning which will affect chick growth, and whether the results previously obtained are affected by using glucose and flour obtained from different sources.

Purposes and Objectives

The purposes of this study are to determine whether various levels of browning of a chick feed composed of bread ingredients affect chick growth, to compare the effect of using different sources of glucose and flour, and to determine the nitrogen and fiber percentages of the feed and chick feces.

Specific objectives of the study are as follows:

1. To determine the effect of browning on chick growth by feeding rations prepared from regular bread ingredients

that have been browned for 0, 5, 10, 15, and 20 minutes at 350°F (176.7°C).

2. To determine the effect on chick growth of the unbrowned feeds when a different source of sugar (glucose) is used.

3. To determine the effect on chick growth of the unbrowned feeds when a different source of flour is used.

4. To determine the percentage of fiber in the browned and unbrowned feeds and chick feces.

5. To determine the percentage of nitrogen in the browned and unbrowned feeds and chick feces.

Hypotheses

The hypotheses postulated for the study are as follows:

H_1: There will be no significant differences in chick weight gains when the bread-ingredient feed has been browned for 0, 5, 10, 15, and 20 minutes at 350°F (176.7°C).

H_2: There will be no significant differences in chick weight gains when the unbrowned feeds are prepared using J. T. Baker Company U.S.P. dextrose or Cerelose brand dextrose.

H_3: There will be no significant differences in chick weight gains when the unbrowned feeds are prepared using Multifoods flour or Pillsbury bread flour.

H_4: There will be no significant differences in fiber percentage (NDF, ADF, or lignin) of feeds or feces caused by browning level, glucose source, or flour source of the feeds.

H_5: There will be no significant differences in nitrogen percentage (NDF, ADF or lignin) of feeds or feces due to browning level, glucose source, or flour source of the feeds.

Assumptions and Limitations

The assumptions made are as follows:

1. Chicks used will be from the same hatching.
2. Chicks will be allowed to eat ad libitum.
3. The experiment steps, including feeding chicks, sample collection, and nitrogen and fiber analyses of the feeds and chick feces, will be done under controlled environmental conditions.
4. Chicks used will vary in weight within a 30 grams range before starting feeding, and not all chicks within a single pen will gain at the same rate.

The limitations accepted for this study are as follows:

1. Only two sources of glucose sugar will be used:
   a. Cerelose, a glucose monohydrate manufactured by Corn Products Company, (Englewood Cliffs, NJ).
   b. U.S.P. dextrose monohydrate, manufactured by J. T. Baker Chemical Company, (Phillipsburg, NJ), obtained from the Oklahoma State University chemical supply room.
2. Only two brands of flour will be used:
   a. Multifoods brand flour, bleached and enriched.
   b. Pillsbury bread flour, unbleached and enriched.

Definition of Terms

The following terms will be used in this study:

1. Essential amino acids -- Amino acids that cannot be synthesized in the body and must be obtained from foods (Robinson and Lawler, 1977).

2. Limiting amino acids -- The essential amino acids that are in short supply in the incomplete protein. If the only source of protein in the diet is an incomplete protein, the one or more essential amino acids inadequately supplied would be the first ones to be used up from the amino-acid pool. After which, the inadequately supplied essential amino acids would limit protein synthesis in the body (Reed, 1980; Guthrie, 1983).

3. Reducing sugar -- A sugar with carbonyl groups having aldehydic or keto carbons. All monosaccharides and some polysaccharides have the ability to reduce an alkaline solution of cupric ions without first undergoing alcohol hydrolysis and are said to be reducing sugars (Campbell, Penfield, and Griswold, 1979; Baum, 1978).

4. Glucose -- A white, crystalline, solid, six-carbon monosaccharide (sugar) soluble in water and insoluble in most organic liquids, found in nature, along with fructose, in many fruit juices. Dextrose is another name for glucose. Chemically, glucose has an aldehydic group that participates in reducing reactions. As the sugar in blood, it is the major source of cellular energy. The body has the ability to convert most other sugars and carbohydrates to glucose.

Parts of protein and fat molecules can also be converted to glucose in the body (Sackheim and Schultz, 1969).

5. Monohydrate -- A compound with one molecule of water that is loosely bound to each molecule of the compound forming a hydrate (Routh, Eyman, and Burton, 1969).

6. U.S.P. dextrose -- U.S.P. is an abbreviation for United States Pharmacopeia, denoting a standard for chemicals and chemical products. Dextrose is another name for glucose being an older term and derived from the fact that a pure solution of this sugar will rotate the plane of polarized light to the right or clockwise (Hawley, 1977; Baum, 1978).

7. Maillard reaction (non-enzymatic browning reaction) -- The amino-groups in proteins when heated react with reducing sugars to form a brown, insoluble, and enzyme-resistant substance with the physical properties of lignin (Lang, 1970; Van Soest, 1978 and 1982).

8. Dietary fiber (cell wall) -- The total fiber in vegetable feedstuffs that cannot be hydrolyzed by enzymes in the digestive tract. Total fiber is a mixture including hemicellulose, pectin substances, gum, mucilages, cellulose, lignin, and glycoprotein. The composition and quantity of these components varies widely from plant to plant (Van Soest, 1978; Bailey, Chesson and Monro, 1978; Schaller, 1978; Cummings, 1976).

9. Neutral detergent fiber (NDF) -- A method for estimation of cell wall constituents in vegetable foodstuffs.

This method utilizes a mild detergent solution for separating the parts of a feed which are nutritionally available in the digestive tract from the other parts which are completely unavailable except through microbial fermentation. Pectin, silica, and tannin may, however, be lost in the NDF method (James and Theander, 1981; Southgate, 1976a).

10. Acid-detergent fiber (ADF) -- An acid-detergent extraction method for analysis of dietary fiber. This method is most effective for determination of the cellulose and lignin components (Southgate, 1976a).

11. Lignin -- A major component of the plant cell wall; it gives added structure to the plant. It is a complex aromatic polymer and is responsible for much of the resistence of the cell wall to chemical degradation and enzymatic digestion (Southgate, 1976b; Hartley, 1978).

CHAPTER II

REVIEW OF LITERATURE

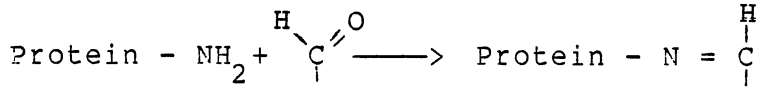
Importance of Maillard Reaction in Food Science and Nutrition

Most food preparation and processing methods such as baking, frying, blanching, pasteurization, or sterilization, require heat. In addition, heating of foods is important for the flavor, color, palatability and appetizing aroma it imparts (Rice and Beuk, 1953; Baldwin et al. 1951).

Chemical Reaction

Since the early 1900's, researchers have observed that foods which are heated or stored for long periods may become brown in color and lose quality and acceptability (Lang, 1970; Rice and Beuk, 1953). In 1912, Maillard first described the heat-induced reaction of glycine (as a single amino acid) with glucose (a reducing sugar) which formed a brown product. From 1913 to 1917, Maillard continued to study the reactions of single amino acids with monosaccharide solutions under different conditions. The reactions occurred when the aldehyde or ketone groups of reducing sugars combined with amino acids, peptides, and proteins

during heating or storage. This reaction was named the "Maillard reaction" (Lang, 1970; Adrian, 1974; Van Soest, 1982; Song, Chichester, and Stadtman, 1966; Mauron, 1981; Saltmarch and Labuza, 1982; Rice and Beuk, 1953). The simplest Maillard reaction was described as follows (Lowry and Thiessen, Jr., 1950):



Because the Maillard reaction in foods could occur without direct enzyme reaction during storage or heating, it was also called the non-enzymatic browning reaction. During and after World War II, many food processing methods were developed and expanded, thus increasing the importance of the Maillard reaction in long-term storage and heat processing of foods (Saltmarch and Labuza, 1982; Feeny and Hill, 1960).

Hodge (1953) found that there were several different pathways that could occur in the Maillard reaction (Figure 1). According to Hodge (1953), the Maillard reaction took place in three stages. In the first stage, the reaction of the free amino group of an amino acid, peptide, or protein condensed with the carbonyl group of a reducing sugar to produce a product known as a Schiff base and water. After that, the reaction of Amadori rearrangement occurred. Both of the compounds were colorless. In the intermediate step, the amino groups were removed from the reducing sugar complex. Subsequently, sugar degradation, fragmentation, and amino

acid degradation occurred depending on various conditions such as temperatures or moisture levels. The compounds might be colorless or yellow. In the third stage, the products from the second step of the reaction became the soluble and insoluble melanoidins or brown compounds in the Maillard reaction (Lang, 1970; Van Soest, 1982; Adrian, 1974; Saltmarch and Labuza, 1982).

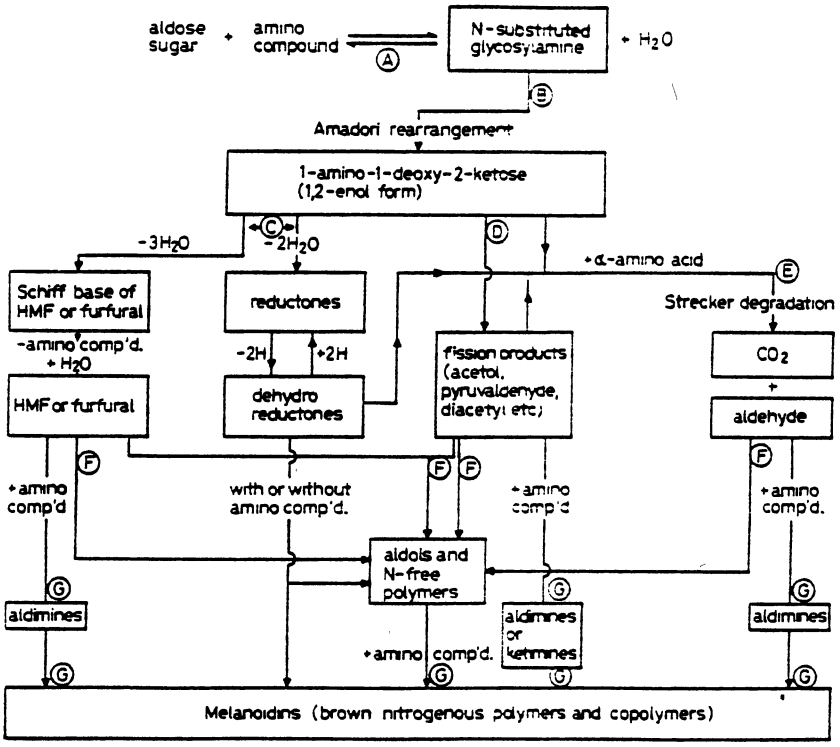


Fig. 1. Scheme of the Maillard reaction (Hodge, 1953).

Color and Flavor

Maillard reactant intermediates through different pathways can contribute a color to foods. In some cases, they give unwanted color as in burned toast (Baltes, 1982); but usually, the Maillard compounds produced by baking, frying, etc., result in a brown, desirable color. The browned crust of a baked product is a good example of this browning reaction (Feeny and Hill, 1960). The color is an apparent indication of the occurrence of the Maillard reaction.

In 1967 Hodge, as reported by Rohan (1970), concluded that much of food flavor also is produced by the degradation of amino acid and reducing sugars in the Maillard reaction. The sulphur-containing amino acids, methionine and cystic acid, condense with reducing sugars and contribute to beef flavor. Similar flavor precursors may contribute to the flavors in bread, coffee, chocolate, peanuts, etc. But in some cases, powdered foods containing carbohydrates and amino acids (dried milk for example) stored in warm, moist conditions, may turn a yellow-to-brown color and develop off-flavors because of the Maillard reaction (Baltes, 1982; Rohan, 1970).

Loss of Nutrients

In the Maillard reaction, not all of the amino acids included in a protein chain are affected. The reducing sugar combines first with the unbound functional nitrogen groups of

the chain causing significant losses of the basic amino acids such as lysine. The epsilon amino group of lysine appears to be the first amino acid to react with reducing sugars to form an amino-sugar group, followed by the sulfur-containing amino acids such as cystine, methionine, and sometimes tryptophan (Adrian, 1974). All of these are essential amino acids for humans; and lysine is a limiting amino acid in many foods, especially in cereal grains. This fact causes some nutritionists to be concerned that the Maillard reaction decreases availability of many essential amino acids (Boctor and Harper, 1968).

Lento, Jr., Underwood, and Willits (1958), observing the Maillard reaction in several different amino acids in glucose solutions at pH 8, heated in sealed tubes at 114^{o}C and 10 p.s.i., reported different levels of brown color development. The amino acids with the amino group in the alpha position formed less color than amino acids with the amino group in the terminal position or those containing diamino groups. Lysine, a simple 6-carbon diamino acid resulted in a greater color development than any of the other amino acids tested.

Sgarbieri et al., (1973a) investigated egg albumin incubated with glucose at 37^{o}C and 70 percent relative humidity for 30 days. The availability of nine essential amino acids in egg albumin was determined by microbiological and chromatographic analyses after browning. The availability of all the amino acids was reduced to various levels. Especially affected were lysine, arginine, and histidine.

The Effect on Gastrointestinal Functions in Animals

The Maillard complex, a product of the Maillard reaction, is a brown, indigestible, and insoluble product. Many investigators have studied the Maillard polymers of various proteins reacting with carbohydrates in animal feed and noted a decrease in the animals' food intake, digestibility, and availability of the essential amino acids (Chichester, 1973; Knipfel, 1975 and 1981).

Most of the peptides are digested by the trypsin, carboxypeptidase, and chymotrypsin enzymes in the intestine. Hansen and Millington (1979) investigated the effect of carboxypeptidase-B enzyme on a solution of poly-L-lysine and glucose. When poly-L-lysine was heated with glucose at 101^{o}C for 60 minutes, and then incubated with carboxypepidase-B at 37^{o}C for 24 hours, only 12 percent of the lysine from the poly-L-lysine-glucose mixture was released. However, 62 percent of the lysine was released by the enzyme as the unheated mixture.

Porter and Rolls (1971) stated that highly digestible proteins were easily utilized by the body, being largely broken down and absorbed from the gut shortly after feeding. The poorly digested proteins such as Maillard proteins, however, were found intact in the gut. It appeared that the Maillard products were poorly absorbed from the small intestine.

Valle-Riestra and Barnes (1970) used radioactivity recovery studies to observe the Maillard residues of lysine-labelled egg white heated with glucose in the different segments of the gut at 2, 7, and 16 hours after feeding test meals to rats. Indications were that the digestive enzymes released less lysine from the Maillard reactants than from heated pure egg white. Radioactivity recovered in the feces showed that much of the Maillard protein resisted digestion and was excreted in the feces. Examination of radioactivity in expired air and urine also showed higher activity in the urine, following ingestion of the labelled Maillard products. This indicated that the lysine or some lysine derivative may have been absorbed from the intestine but could not be utilized and was, therefore, excreted in the urine. Similar results were reported by Perkins, Baker, Johnson, and Makowski, (1981). The microflora in the large intestine played an important role in attacking nitrogenous substances such as urea, amides, amino acids, and proteins. When the Maillard products accumulated in the large intestine, they may have been degraded by the action of the microflora and absorbed. The radioactivity of the Maillard proteins was then measured in the urine of the rats, showing that the compound was probably absorbed from the large intestine and largely excreted in the urine without being metabolized (Tanaka, Lee and Chichester, 1975; Sgarbieri, Amaya, Tanaka, and Chichester, 1973b).

The Effect of the Maillard Reaction on Animal Growth

Since Maillard complexes, such as bound lysine and other derivatives, are digestive-enzyme resistant (or only slowly hydrolyzed), the amino acids are not liberated, and protein synthesis is less efficient. Buraczewski, Buraczewska, and Ford in 1967, as reported in Porter and rolls (1971), showed that "unavailable" peptides in the intestinal contents of rats accumulated at the absorption sites and prevented the uptake of amino acids.

Johnson, Baker, and Perkins (1977), studied Maillard bound fructose-phenylalanine as a source of phenylalanine for chicks receiving a phenylalanine-deficient diet for six days. It was shown that chicks fed an equimolar quantity of phenylalanine in the form of the fructose-phenylalanine did not have as significant weight gains as chicks fed pure crystalline phenylalanine. It appeared that fructose-phenylalanine was nutritionally unavailable to the chick.

Knight and Hanson (1982, 1983) have studied two feeding trials using chicks. In Trial 1, a chick feed was prepared using glucose (dextrose manufactured by J. T. Baker Chemical Company), Pillsbury bread flour, egg solids, yeast, and oil. Portions of the feed were browned for 10, 20, and 30 minutes at 350^{o}F (176.7^{o}C). After feeding chicks for seven days, chicks fed the 20-minute browned ration gained little weight, and chicks fed the 30-minute browned mixture showed no growth

at all. However, after supplementing the browned feed with lysine, the chicks began to grow normally.

In Trial 2, using a different source of glucose (Cerelose from Corn Products Company) and Multifoods flour, the feeds were browned at 350°F (176.7°C) for 0, 5, 10, 15, and 20 minutes. After seven days, chicks fed the 15-minute browned ration showed the best growth. There were no significant differences among weight gains of chicks fed the 0, 5, and 10 minute browned feeds. Chicks given the 20-minute browned ration experienced the least weight gain of all treatments, but gained more than chicks on the 20-minute browning treatment in Trial 1.

Kimiagar et al. (1980) observed the effect of Maillard browned egg albumin with glucose fed to rats for 12 months. One month into the study, there were no significant differences in weight gain, serum components, or tissue enzyme activities between the control and experimental groups. In the second and following months, the animals on the browned diet showed a slower rate of gain, higher blood urea nitrogen, lower hemoglobin and hematocrit levels, and enlargement of some organs. The Maillard proteins evidently caused nutritive losses and affected other physiological conditions in the long-term feeding that were not found in the short-term. Similar research has been subsequently reported by others (Lee, Kimiagar, Pintauro, and Chichester, 1981; Lee, Pintauro, and Chichester, 1982).

Factors Affecting the Maillard Reaction

The rate and extent of Maillard reactions are affected by time and temperature, types and amounts of the reactants, pH levels, and moisture content (Lang, 1970).

Time and Temperature

In reviewing nutrient damage in the Maillard reaction, Lang (1970) concluded that damage is proportional to time and temperature. Some investigators he reviewed observed that reacting casein with glucose for three minutes at 121°C (249.8°F) reduced the protein efficiency about 25 percent in rats, and reacting casein with glucose for 15 minutes at 121°C (249.8°F) caused severe protein damage. The rats given this mixture lost weight.

Frazier et al. (1953) fed fibrin-glucose mixtures dry-heated at different temperatures for 30 minutes to rats for seven days. The rat weight gains, nitrogen intakes, and absorbed nitrogen values were less as the temperatures increased, but fecal nitrogen increased.

The Effect of Reactants

Various types and amounts of amino acids and proteins affected the rate and extent of the Maillard reaction. Knipfel (1975) also observed that using different types of protein, such as casein, soy, or egg, when combined with glucose and autoclaved, affected the total nutritional status of

rats. A mixture of egg protein and glucose produced smaller weight gains and food intakes than did the mixtures of casein or soy heated with the glucose. Also, the net protein ratios (NPR) of the egg-glucose and the soy-glucose mixture were similar but lower than the NPR's of the casein-glucose mixture.

Different types and amounts of sugar combined with the same proteins or amino acids also caused different effects. The sugars with shorter carbon chains caused an increased rate of reaction. Pentoses were the most reactive, followed by hexoses, disaccharides, and polysaccharides. The heated mixtures of hexoses with lysine caused an average 42 percent loss of the amino acids, whereas pentoses caused an average 66 percent loss (Adrian, 1974).

Frazier et al. (1953) investigated the reaction of fibrin with glucose, fructose, sucrose, and cornstarch when heated. The mixtures of glucose-fibrin heated at 302^{o}F (150^{o}C) for 30 minutes and fructose-fibrin heated at 266^{o}F (130^{o}C) caused protein-damage and affected the growth of rats. Although sucrose was not a reducing sugar, it appeared to have undergone inversion to glucose and fructose at higher temperatures, 366.8^{o}F (186^{o}C), since the concentration of reducing compounds and the reaction with amino acids increased with increasing temperatures. Cornstarch, even at temperatures of 410^{o}F (210^{o}C), did not decompose to reducing components or react with amino acids. Pomeranz, Johnson, and Shellenberger (1962) also studied the effect of various

sugars on browning. They showed that sugars without reducing groups did not brown as completely and had little reactivity.

Karel and Labuza (1968) when studying a model system which contained sucrose and organic acid noted the development of non-enzymatic browning even at a low moisture content. Hurrell and Carpenter (1977) fermented a mixture of sucrose with yeast and combined it with a high-protein cake containing ovalbumin and lactalbumin, which was then baked and toasted. The results showed that the heated mixture had severe fluorodinitrobezene (FDNB)-reactive lysine loss and protein damage when fed to rats. However, a similar albumin-sucrose mixture that was baked and toasted without prior fermentation caused little reduction in lysine and no nutritive failure to the rats. It appeared that in both studies, the sucrose was hydrolyzed to molecules of fructose and glucose, thus increasing the concentration of reducing sugars and participation in the Maillard reaction.

The Effect of pH Levels

Frazier et al. (1953) autoclaved 5 percent glucose-amino acid solutions at different pH levels at 240^{o}F (115.6^{o}C) for one hour. The solution mixture at pH 7 had less nutritive damage than did the pH 9 and 10 solutions as determined by rat repletion assay. Others investigated the amino acids, lysine, glutamic acid and DL-alanine. Amino acids were reacted with a 0.025M glucose solution at different pH values

with an autoclave temperature of 114°C (237.2°F) for 20 minutes. The level of browning increased as the pH level increased (from pH 3 to pH 9), and the lysine-glucose solution had more browning than the other two amino acid solutions (Willits, Underwood, Lento, Jr., and Ricciuti, 1958).

A number of studies showed that the rate and degree of the Maillard reaction increased proportionally to pH levels, from pH 3 up to pH 8 (Adrian, 1974; Saltmarch and Labuza, 1982). Adrian stated the lesser reaction at lower pH might be the reason that heated meat was more stable in nutritive value than heated fishmeal (Adrian, 1974).

The Effect of Moisture

Schroeder, Iacabellis, Lees, and Smith (1953), determined that the nutritive value of the milk proteins in the Maillard reaction was affected by water. Dried skim milk reconstituted with water to levels of 3.5 (normal milk reconstitution), 10.5, 17.5, or 24.5 percent protein was autoclaved. Crystalline trypsin or chymotrypsin was used to determine the amino nitrogen released. Water increased the protein efficiency of the dried skim milk and whole milk powders, even at the minimum water level of 24.5 percent protein (seven times the normal reconstitution). The water apparently prevented the nutritive value loss which generally occurred in the dry condition.

Lea and Hannan in 1949 and 1952 demonstrated, as Adrian (1974) reviewed, that the Maillard reaction did not happen in

the strictly anhydrous state. The greatest Maillard reaction occurred between a relative humidity of 40-70 percent. As water dilution increased, the rate of Maillard reaction was decreased. The Maillard reaction also did not occur in a very diluted solution (Adrian, 1974).

Several factors were proposed to explain the apparently contradicting relationship between the Maillard reaction and water. In the browning reaction, water was produced in the initial glycosylamine reaction. Foods which contained water may have further diluted the reactive components. On the other hand, the rate of Maillard reaction might have been increased due to increased water content which sped up the mobility of the reactive compounds. If the dilution effect overcame the faster mobility of the reactive components, the overall rate of the Maillard reaction would be decreased (Saltmarch and Labuza, 1982).

The Relationship of Fiber and Maillard Reaction

The apparent dietary fiber content of food can be increased by Maillard compounds which have the physical and chemical properties of lignin but have a high nitrogen content. These kinds of materials are called "artifact lignin" (Van Soest, 1965, 1978 and 1982; Cummings, 1978; Van Soest and McQueen, 1973). Table I shows the effect of different heat treatments on the content of acid detergent fiber, lignin, and insoluble protein.

TABLE I

EFFECT OF DIFFERENT TREATMENTS UPON THE YIELD AND COMPOSITION OF THE ACID-DETERGENT FIBER OF FRESH ORCHARD-LADINO MIXTURE
(from Van Soest, 1965)

Moisture Content[a], %	Heat Treatment			Fiber %[b]	Insol. Protein (N X 6.25), %[b]	Apparent Lignin %[b]
	Temp., °C		Time, hr			
9	100	dried in open dish	20	32.4	2.2	7.0
9	100	dried in open dish	44	33.1	2.5	7.5
9	100	dried in open dish	120	34.6	3.4	9.3
9	100	$+H_2O$ in stoppered flask[c]	20	41.8	8.3	14.4
9	100	2 g boiled in 100 ml H_2O	20	41.1	9.3	15.9

a Moisture content of the starting material.
b Expressed as percentage of whole forage dry matter.
c Equivalent to 40% moisture.

TABLE II

COMPOSITION OF TOASTED PRODUCTS

Sample	ADF	ADF N x 6.25	ADF N/total N	Crude Lignin
		Dry Basis %		
Untoasted bread	0.6	0.1	0.7	0.2
Toast	1.5	0.6	3.7	0.8
Potato chips	4.1	0.6	9.0	0.9
Tortilla chips	2.7	0.7	7.8	1.5
Corn Flakes	3.9	2.5	31.0	1.8
Wheat Chex	6.5	3.8	32.0	3.5

Breakfast cereals such as Wheat Chex, corn flakes and other browned products have a significant amount of this "artifact lignin" (Van Soest, 1978). This phenomena is illustrated in Table II (Van Soest, 1978).

Because the amount of dietary fiber seems to increase with heating due to the Maillard products formed, those products might also have other characteristics of dietary fiber such as water-holding capacity, bulk density, ion-exchange capacity and bacterial fermentability, which have important nutritional considerations (Van Soest, 1978; Eastwood and Mitchell, 1976; Lewis, 1978).

Maillard reactants are considered to be "artifact lignin" and are quantitatively recovered in the residues from neutral-detergent extraction, acid-detergent extraction,

or in the crude lignin (Van Soest, 1963, 1968, and 1982; Van Soest and McQueen, 1973). A problem in the determination of the neutral-detergent (NDF) or acide-detergent fiber (ADF) can be filtration. Some foodstuffs contain high amounts of protein which can interfere with filtration. Sodium sulfite is used in the neutral-detergent fiber procedures to attack disulfide bridges and the linkages between the aromatic compounds in the protein chains. On the other hand, sodium sulfite may cause the loss of lignin subunits and also attack the Maillard polymers which affects the determination of neutral-detergent fiber lower (James and Theander, 1981). When the foodstuffs contain high contents of starch, starch gels may form that will not filter through the crucibles used in neutral-detergent fiber analysis. α-amylase may be used to hydrolyze the starch before neutral-detergent fiber analysis (Schaller, 1978; James and Theander, 1981).

The Maillard polymers are mainly recovered in the lignin fraction and are insoluble in 72 percent sulfuric acid. Determination of lignin with Maillard polymers by the acid-detergent lignin procedure appears to be the optimal method. In this method, acid-detergent fiber is treated with 72 percent sulfuric acid to separate polysaccharides from lignin. Then the determination of lignin is obtained by gravimetry, also substracting ash weight correct for inorganic substances such as minerals of dietary fiber (Van Soest, 1982; Hartley, 1978).

Summary of Review of Literature

Most food preparation and processing methods require heat. Heating of food is important for flavor, color, palatability, and appetizing aroma. In some cases, heating food gives unwanted color and flavor. Researches have found that when reducing sugars are heated with amino acids such as lysine, a brown, insoluble, and indigestible product is formed. This reaction is called the Maillard reaction. The results of several studies show that Maillard compounds resist digestion and are excreted. The growth of experimental animals is adversely affected due to the unavailability of the Maillard-bound proteins. Several factors affect the rate and extent of the Maillard reaction such as temperatures and time, types and amounts of the reactants, pH levels, and moisture content.

Although the content of dietary fiber is apparently increased by the Maillard reaction, the exact role of the Maillard products as dietary fiber has not been determined.

CHAPTER III

METHOD AND PROCEDURES

A review of related literature indicated that an amino-sugar binding group in the Maillard reaction appeared to decrease the utilization of the nutrients and thus affect chick growth. Knight and Hanson (1982, 1983) studied two feeding trials, Trial 1 and Trial 2, using chicks. Since neither of these trials clearly established the level of browning where growth is retarded, it was necessary to repeat Trial 2. The effects that the different sources of flour and glucose may have had on the results obtained in Trial 1 and Trial 2 also needed to be determined. To discover whether or not the browned products had the characteristics of fiber and increased fecal nitrogen, fiber and nitrogen in the feeds and chick feces was compared.

The purpose of this study was two-fold. First was the determination of the effects of browning chick feeds on various physiological parameters of growing chicks. Chick feeds made from the same source of flour and sugar and browned for five different time periods were fed, and the growth rate determined. Second was determination of the effect of unbrowned feeds made from different sources of sugar and flour on the growth rate of chicks. Fiber and nitrogen

content in the feeds and chick feces was determined in both feeding trials, i.e., the one using browned feeds and the one using unbrowned feeds. The topics included in this chapter are research design, feed preparation, chicks, growth data and fecal collection, procedures for fiber and nitrogen analyses, and statistical analyses.

Research Design

A completely randomized design was applied to this experiment. Eight different kinds of chick rations were considered as eight different treatments or independent variables.

Feeds were randomized for the 17 cages (pens), and the pens were the experimental units. The numbers of pens for each feed treatment (n) were not the same; i.e., Treatments 0, 5, 10, 15 and 20 had two pens; Treatment I had three pens; and Treatment II_1 and Treatment II_2 had two pens.

The chicks were randomized and assigned to 17 pens in number order, so they were considered nearly homogeneous. The number of chicks fed per treatment was determined by amounts of feed available. The chicks in each pen were fed the assigned ration for seven days. The chicks were weighed individually, but feed intakes and fecal weights were determined per pen.

Experiments performed were conducted under controlled environmental conditions in order to test the hypotheses given in Chapter I.

Feed Preparation

Eight feed treatments were prepared. Five (Treatments 0, 5, 10, 15 and 20) were made of flour and glucose from the same source, the only variation being the length of time that they were browned. The other three (Treatments I, II_1 and II_2) were made using flour and sugar from different sources and were left unbrowned. The composition of the basal chick ration is given in Table III. The sugars and flours used in the different feeds are given in Table IV.

TABLE III

COMPOSITION OF THE BASIC FEED

Ingredients	% of ration
Flour (Pillsbury bread or Multifoods flour)	47.80
Sugar (USP Dextrose or Cerelose)	10.00
Egg solids	26.40
Yeast	5.00
Vegetable oil (Wesson)	5.00
*Mineral salts	5.37
*Vitamins	0.40

* Compositions of mineral salts and vitamins are given in Appendix (Tables XXXIII and XXXIV).

TABLE IV

GLUCOSES AND FLOURS USED IN THE DIFFERENT FEEDS

Treatment	Cerelose Sugar	USP dextrose sugar	Pillsbury bread flour	Multifoods flour
0	*			*
5	*			*
10	*			*
15	*			*
20	*			*
I		*	*	
II_1	*			*
II_2	*		*	

The first five chick feeds, Treatments 0, 5, 10, 15 and 20 (Table IV) were prepared using Cerelose sugar and Multifoods flour left over from Trial 2, performed by Knight and Hanson (1983), and were browned for 0, 5, 10, 15 and 20 minutes. Treatment I was unbrowned feed using J. T. Baker Company U.S.P. dextrose and Pillsbury bread flour as used in Trial 1 (Knight and Hanson, 1982). Treatment II_2 feed was also unbrowned feed using Cerelose sugar as used in Trial 2 and Pillsbury bread flour as was used in Trial 1. Treatment I and Treatment II_2 feeds were compared with

Treatment II_1 feed which had the same composition, i.e., Cerelose sugar and Multifoods flour, as were used in the browned feeds.

The feeds for all treatments were prepared by combining the flour, sugar, vegetable oil, yeasts and egg solids and mixed thoroughly. For the treatments requiring a browned feed, the oven was preheated to 350°F (176.7°C). The feed mixtures were placed in shallow pans filled to a uniform depth and heated for the appropriate time intervals. The five-minute browned feed mixture was left in the oven five minutes, removed, mixed uniformly, and the internal temperature of the feed in each pan taken. The average temperature of this browning level was 179.6°F (82°C). The feed was then spread in large trays to facilitate cooling. Feeds browned for ten minutes were put into the oven for two five-minute intervals. Between the two five-minute intervals, the mixture was taken out of the oven and mixed uniformly. After the second five minutes, it was again removed, mixed uniformly, and the temperatures recorded. The average temperature for this browning level was 194°F (90°C). The procedure for browning feeds for fifteen and twenty minutes was the same as for the five and ten minute brownings. The feed browned fifteen minutes was put in the oven for three five-minute intervals, and the twenty-minute browned feed was heated for four five-minute intervals. The average temperature of the mixtures, taken upon final removal, was 208.4°F (98°C) for the fifteen-minute browned feed, and 230°F (110°C)

(110°C) for the twenty-minute browned feed. After all feeds had cooled to approximately 80°F (26.7°C), the feed for each treatment, browned and unbrowned, was weighed and the correct percentages of the mineral salts and vitamins meeting the growth requirements for chicks were added and mixed well. All the chick feeds were stored in the freezer until used.

Chicks

Chicks are more sensitive to lysine for their growth than most other laboratory animals and are appropriate for lysine assay or lysine binding studies (Fisher, 1974). The chicks for this research were provided by the Animal Science Department at Oklahoma State University. Seventy-eight 9-day-old chicks were used. Chicks with weights ranging from 110 grams to 140 grams were selected.

For Treatments 0, 5, 10, 15 and 20, there were three chicks per pen and two pens per treatment (Table V). The number of chicks per pen and pen numbers for the unbrowned Treatments I, II_1 and II_2 are shown in Table VI. Treatment I had three pens, eight chicks per pen. For Treatment II_1 and Treatment II_2, there were six chicks per pen and two pens per treatment.

Growth Data and Fecal Collection

The feed and water were fed to the chicks ad libitum for seven days. The amount of feed given to the chicks was

TABLE V

THE NUMBER OF CHICKS AND PEN NUMBERS FOR TREATMENTS 0, 5, 10, 15 and 20

Treatment	0		5		10		15		20	
Pen No.	12	15	1	14	6	11	8	10	16	17
No. of Chicks	3	3	3	3	3	3	3	3	3	3

TABLE VI

THE NUMBER OF CHICKS AND PEN NUMBERS FOR TREATMENTS I, II_1, II_2

Treatment	I			II_1		II_2	
Pen No.	3	7	9	4	5	2	13
No. of Chicks	8	8	8	6	6	6	6

weighed and recorded every day. The chicks were weighed individually on days 0, 3, 5 and 7. A 24-hour feces sample was collected on days four and seven for each pen. The feces were dried at 122°F (50°C) overnight in a forced draft oven, and ground to pass through a No. 20 mesh.

Procedures for Fiber and Nitrogen Analyses

Determination of Neutral-Detergent Fiber (NDF) Procedures

Since the samples of feeds had a high starch content, bacterial α-amylase was used as a pretreatment to hydrolyze the starch before determining neutral-detergent fiber. Samples of chick feces weighing 0.5-0.6 grams and samples of chick feed weighing about 1.0 gram were used in determining neutral-detergent fiber. The sample weight was proportional to the volume of α-amylase solution used for pretreatment. The McQueen and Nicholson procedure was used for the pretreatment (McQueen and Nicholson, 1979). Two methods of NDF determinations of feeds and chick feces were used. One of the NDF determinations added sodium sulfite after the pretreatment; the other NDF determination did not add sodium sulfite. After that, both of the NDF determinations of feeds and chick feces were made following the Association of Official Analytical Chemists (AOAC) approved methods (Van Soest and Wine, 1967).

Determination of Acid-Detergent Fiber (ADF) and Lignin Procedures

Chick fecal samples weighing about 1.0 gram and samples of chick feeds weighing about 2.0 grams were used for determining acid-detergent fiber. The chick feeds were subjected to α-amylase pretreatment. The sample weight of chick feeds was proportional to the volume of α-amylase solution used for the pretreatment. The McQueen and Nicholson procedure was used for the pretreatment (McQueen and Nicholson, 1979). The ADF determinations of chick feces and chick feeds were made using the approved AOAC method (Van Soest, 1963). The acid-detergent fiber test was also used as a preparatory step for acid-detergent lignin procedure. Lignins were determined using the acid-detergent lignin procedure (Van Soest, 1963).

Determination of Nitrogen Procedure

The percentage of nitrogen in the feeds and chick feces were determined using automated equipment. The determinations were carried out using a Tecator (Hendon, VA) Digestion System 40 and a Kjeltec Auto 1030 Analyzer.

Statistical Analyses

Data were analyzed by using the Statistical Analysis System (SAS, 1979). Covariance analysis was used to adjust weight gains of the chicks depending on feed intake,

enabling the estimation to the effect of certain feed treatments on these weight gains (Steel and Torrie, 1960). Feed intake of the chicks and fiber and nitrogen percentages in the feeds and chick feces were determined by analysis of variance (ANOVA). The Duncan Multiple Range Test was used to test the significance of the differences among means for the data at the 0.05 level of significance (Snedecor and Cochran, 1973).

CHAPTER IV

RESULTS AND DISCUSSION

This study was done in order to determine how a chick feed using the same source of sugar and flour at five browning levels and a chick feed using different sources of sugar and flour without browning affected the growth rate of chicks and the fiber and nitrogen percentages of the feeds and chick feces. The feed intake and chick growth were recorded. The fiber and nitrogen percentages in the feeds and feces were performed objectively.

Evaluation of Seven-Day Feed Intake

The color of chick feeds changed from light brown to medium brown as the browning levels of the feeds increased. The results show (Table VII) that the feed intake of chicks decreased as the browning levels of the feeds increased with the exception of Treatment 15. The feed intake of chicks for Treatment 15 was greater than for the browned treatments 10, and 20. Feed intake of chicks for Treatment 20 was less than for the other treatments. Feed intake of chicks for the unbrowned treatments was greater than for the browned treatments.

TABLE VII

THE MEAN VALUES OF FEED INTAKE OF THE CHICKS GIVEN DIFFERENT TREATMENTS FOR SEVEN DAYS

Treatments	II_2	I	II_1(0)*	5	10	15	20
Average Daily Feed Intake (gms)	39.6	39.4	37.4	36.2	30.1	35.8	21.5

* Treatment II_1 and Treatment 0 were the same feed blend. Since Treatment II_1 and Treatment 0 were composed of the same feed blend, the feed consumption results from these four were pooled.

Many investigators have studied the Maillard polymers of different proteins which form when reacted with carbohydrates in heated animal feeds and discovered a decrease in the animal's intake and the digestibility and availability of the essential amino acids in the feed (Chichester, 1973; Knipfel, 1975). Others have reported that the formation of color and flavor in the Maillard reaction contributed appetizing aroma and palatablity of the feed and thus encouraged feed intake (Feeny and Hill, 1960; Rohan, 1970). In this study it might be that the color and flavor of feed from Treatment 15, resulting from the Maillard reaction, attracted the chicks and increased their feed intake. Another, and perhaps more likely explanation of the increased intake of Treatment 15 feed, would be that

moderate heating does appear to improve the digestibility and acceptance of starch (Lang, 1970).

According to the results of analysis of variance, there were significant differences among feed intakes of chicks fed different treatments (Table XXVII. Appendix). Table VIII shows that the feed intake of chicks fed Treatment 20 was significantly less than chicks fed other treatments.

The plots of chick weight gains for different treatments, unbrowned and browned, are given in Figure 2. Table IX and Figure 2 show that in the study of the browned Treatments $II_1(0)$, 5, 10, 15, and 20, chicks fed Treatment 15 gained weight best, and chicks fed Treatment 20 gained less weight than those fed the other treatments. Table IX and Figure 2 show that in the study of the unbrowned Treatments $II_1(0)$, II_2, and I, chicks fed Treatment I gained less weight than chicks fed Treatments II_2 and $II_1(0)$. Chicks fed Treatment $II_1(0)$ gained more weight than those fed Treatment II_2. Table IX and Figure 2 also show that chicks fed Treatment 15 had the best growth among both unbrowned and browned treatments. Chicks fed Treatment 20 gained less weight than all other treatments. Chicks fed unbrowned treatments, however, gained more weight than chicks fed the browned Treatments 5, 10, and 20.

Evaluation of Chick Growth

Because analysis of variance showed that there are significant differences among feed intakes of chicks given

TABLE VIII

THE DUNCAN MULTIPLE RANGE TEST FOR FEED INTAKE OF CHICKS GIVEN DIFFERENT TREATMENTS ($\alpha = 0.05$)

Treatment	No. of Pens	Mean (gms)*	Duncan Grouping
II_2	2	39.560	A
I	3	39.395	A A A
$II_1(0)$	4	37.407	A A A A
5	2	36.193	A A A A
15	2	35.755	A B B B
10	2	30.078	B B
20	2	21.514	C

* Means with the same letter are not significantly different.

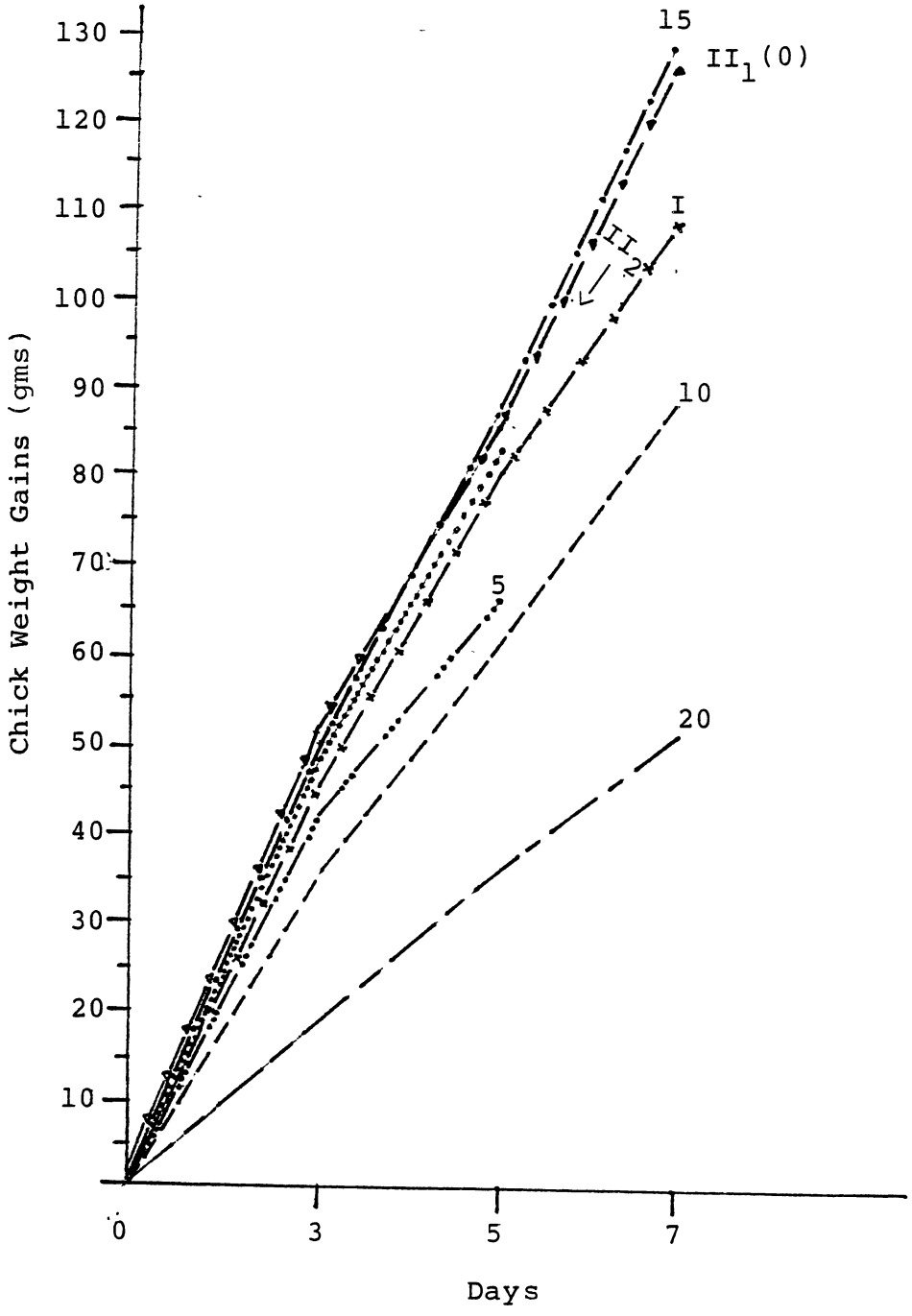


Figure 2. The plots of chick weight gains vs. days on unbrowned and browned treatments.

TABLE IX

CHICK WEIGHT GAINS FOR EACH TREATMENT
AT DIFFERENT PERIODS AND AT
DIFFERENT CHICK AGES

Treatments / Weight gains at period intervals / Chicks ages in days	[a] II_2		[a] I		[b] II_1	[b] 0	[a] 5		[a] 10		[b] 15	[b] 20
	9	10	9	10	9	10	9	10	9	10	10	10
Day 0 to Day 3	49	48	50	42	54	50	28	57	36	38	50	19
Day 0 to Day 5	81	85	85	77	88	86	60	74	61	63	88	37
Day 3 to Day 5	32	37	35	35	34	36	32	17	25	25	38	18
Day 0 to Day 7	[c]	[c]	114	103	128	123	[c]	[c]	87	90	128	51

a = The wrong feed (a standard chick feed, was accidentally put in the feeders from Pen 8 to pen 17 on the second day (Day 1) of the experiment by personnel not associated with this experiment. Treatments of the chicks from Pen 8 to Pen 17 were, therefore, delayed one day, making them one day older than chicks from Pen 1 to Pen 7.

b = Chick weight gains were averaged for treatments using chicks in different pens that were of the same age.

c = Missing data.

different treatments, covariance analysis was used to adjust chick weight gains depending on feed intakes of chicks to see the difference between treatments. The covariance model considered the observed value of the dependent variable (chick weight gains) affected by the particular treatment levels or groups from which the observation came, and the values of the concomitant variable (feed intake). The equation of the covariance model used is shown as follows:

$$Y_{ijk} = \mu + \gamma_i + \beta_i W_{ij} + \varepsilon_{ijk}$$

Y_{ijk} = Observed value of chick weight gains

μ = Mean of chick weight gains

γ_i = Effect of different treatment levels

$\beta_i W_{ij}$ = Effect of feed intake of chicks

ε_{ijk} = Residual effect

Determination of Chick Weight Gains from Day 0 to Day 3

The homogeneity of slopes ( ) of chick weight gains for seven different treatments at time interval Day 0 to Day 3 was tested. Statistical analysis showed that the slopes of chick weight gains for different treatments were significantly different (Table XXVIII. Appendix). For this reason, the slope value of chick weight gains for each treatment was calculated. The slope value of chick weight gains for each treatment (Table X) shows that the slope values of Treatment I and 20 were significantly different from the slope values of the other treatments.

TABLE X

SLOPE VALUES OF CHICK WEIGHT GAINS
FOR ALL TREATMENTS FROM
DAY 0 TO DAY 3

Treatment	Slope Value
I	-1.1732
II_2	0.0429
$II_1(0)$	0.1435
5	0.5625
10	0.2936
15	0.6461
20	-4.6778

The homogeneity of the slopes (β) of the other treatments (II_2, $II_1(0)$, 5, 10, and 15) was then tested. Statistical analysis showed that there were no significant differences among chick weight gains for treatments II_2, $II_1(0)$, 5, 10, and 15 (Table XXIX. Appendix). Since the slopes of Treatments II_2, $II_1(0)$, 5, 10, and 15 were not significant, the estimation of the slope was calculated (Table XXIX. Appendix). To do this, chick weight gains for each treatment were adjusted in order to estimate the effect of different treatments on chick weight gains. The homogeneity of the treatment effect was tested. Statistical analysis showed that there were no significant differences among

chick weight gains for Treatments II_2, $II_1(0)$, 5, 10, and 15 (Table XXIX. Appendix). Table XI shows that there were no significant differences among Treatments II_2, $II_1(0)$, 5, 10, and 15.

Determination of Chick Weight Gains from Day 0 to Day 5

Testing the homogeneity of slopes showed that there were no significant differences among the slopes (β) of the chick weight gains for different treatments (Table XXIX. Appendix). Testing the homogeneity of treatment effect (γ) showed that there were significant differences among chick weight gains for different treatments (Table XXIX. Appendix). Table XII shows that chicks fed Treatment 20 gained significantly less weight than chicks fed the other treatments. There were no significant differences among Treatments II_2, I, $II_1(0)$, and 15. There were no significant differences between Treatments 5 and 10. The chick weight gains for Treatments II_2, I, $II_1(0)$, and 15 were significantly higher than Treatments 5, 10, and 20.

Determination of Chick Weight Gains from Day 3 to Day 5

Testing the homogeneity of slopes showed that there were no significant differences among the slopes (β) of chick weight gains for different treatments (Table XXIX. Appendix). Testing the homogeneity of treatment effect (γ)

TABLE XI

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF CHICK WEIGHT GAINS ADJUSTED FOR DIFFERENT TREATMENTS FROM DAY 0 TO DAY 3 ($\alpha = 0.05$)

Treatment	No. of Chicks	Mean (gms)*	Duncan Grouping
II_1(0)	18	8.4901	A
			A
15	6	8.1091	A
			A
II_2	12	5.5884	A
			A
5	6	5.1706	A
			A
10	6	4.9247	A

* Means with the same letter are not significantly different.

TABLE XII

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF CHICK WEIGHT GAINS ADJUSTED FOR DIFFERENT TREATMENTS FROM DAY 0 TO DAY 5 (α=0.05)

Treatment	No. of Chicks	Mean (gms)*	Duncan Grouping
II_1(0)	18	12.987	A
15	6	12.853	A A
I	24	11.224	A A
II_2	12	11.045	A A
10	6	8.375	B
5	6	8.331	B B
20	6	4.423	C

* Means with the same letter are not significantly different.

showed that there were significant differences among chick weight gains for different treatments (Table XXIX. Appendix). Table XIII shows that there were no significant differences among Treatments II_2, I, $II_1(0)$, and 15. There were also no significant differences among Treatments 5, 10, and 20. The chick weight gains for Treatments II_2, I, $II_1(0)$, and 15 were higher than for Treatments 5, 10, and 20.

Determination of Chick Weight Gains from Day 0 to Day 7

Testing the homogeneity of slopes showed that there were no significant differences among the slopes (β) of chick weight gains for different treatments (Table XXIX. Appendix).

Testing the homogeneity of treatment effect (γ) showed there were significant differences among chick weight gains for different treatments (Table XXIX. Appendix). Table XIV shows that chicks fed Treatment 20 gained significantly less weight than chicks fed the other treatments. There were no significant differences among treatments $II_1(0)$, 15 and II_2. The chick weight gains for Treatments $II_1(0)$, 15 and II_2 were significantly higher than all the other treatments.

Determination of Feed Efficiency

The effects of various treatments on feed efficiency of chicks for all seven days is given in Table XV. Treatment 15 was more efficient than all other treatments, both unbrowned

TABLE XIII

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF CHICK WEIGHT GAINS ADJUSTED FOR DIFFERENT TREATMENTS FROM DAY 3 TO DAY 5 ($\alpha = 0.05$)

Treatment	No. of Chicks	Mean (gms)*	Duncan Grouping
15	6	6.4155	A
$II_1(0)$	18	5.2434	A A A
I	24	3.9298	A B
II_2	12	2.8806	A B A B A B
10	6	1.6317	B B C
20	6	1.2349	B C C
5	6	-1.9362	C C

* Means with the same letter are not significantly different.

TABLE XIV

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF CHICK WEIGHT GAINS ADJUSTED FOR DIFFERENT TREATMENTS FROM DAY 0 TO DAY 7 ($\alpha = 0.05$)

Treatment	No. of Chicks	Meqn (qms)*	Duncan Grouping			
II_1(0)	18	14.705	A			
15	6	14.593	A			
II_2**	12	12.645	A	B		
I	24	11.840		B	C	
5**	6	9.794			C	
10	6	9.605			C	
20	6	5.192				D

* Means with the same letter are not significantly different.

** Weight gains for days 5-7 extrapolated data.

and browned, and Treatment 20 was less efficient than all the other treatments. Of the unbrowned feeds, Treatment $II_1(0)$ was the most efficient.

TABLE XV

THE EFFECT OF DIFFERENT TREATMENTS ON FEED EFFICIENCY OF CHICKS FOR SEVEN DAYS

Treatment	Average Daily Weight gains (gms)	Average Daily Feed Consumption (gms)	Feed Efficiency
II_2	16.59	39.56	2.38
I	15.77	39.39	2.49
$II_1(0)$	18.42	37.41	2.03
5	13.41	36.20	2.69
10	12.61	30.08	2.39
15	18.16	35.76	1.97
20	7.34	21.52	2.93

Discussion of Chick Weight gains

Maillard reactions associated with even mild heating may affect availability of amino acids such as lysine without reducing protein digestibility or food intake greatly. After extensive heating, the unavailability of amino acid increases. Food intake and protein efficiency are also reduced to a greater extent with extensive heating than

with mild heating. Due to decreased food intake, caloric intake might not meet the animal's energy requirements. The lack of sufficient energy would cause inefficient utilization of the protein ingested by the animals (Knipfel, 1981). On the other hand, Maillard complexes (bound amino acids which are digestive-enzyme resistant) will not liberate the amino acid and therefore protein synthesis is less efficient (Porter and Rolls, 1971). Animal growth and maintenance will, therefore, be affected.

Comparing all of the feed intakes of chicks, plots, chick weight gains, and feed efficiencies for different treatments, the results showed that chicks given unbrowned treatments had better weight gains than chicks given browned treatments, with the exception of Treatment 15. Of the unbrowned treatments, chicks fed Treatment $II_1(0)$ gained the most weight, perhaps because of the different brands of glucose and flour used, with the Cerelose glucose/Multifoods flour appearing to give the best growth.

Chicks given Treatment 15 grew the best of all the browned treatments. Chicks given Treatment 20 had some weight gain, but less than all other treatments, browned or unbrowned. Knight and Hanson (1982, 1983) have studied two feeding trials using chicks. The results of Knight and Hanson's Trial 2 showed that chicks fed the 15-minute-browned ration gained the most weight, and chicks fed the 20-minute-browned ration gained the least. However, in the Knight-Hanson Trial 1, using U.S.P. dextrose and Pillsbury

flour, chicks fed the 20-minute-browned ration had no appreciable weight gain.

Denaturation of a protein molecule such as lactalbumin, ovalbumin, or casein by mild heating can enhance its digestibility by proteases. Palatability, digestibility, and availability of certain nutrients of foods of plant origin can be improved by heating for adequate time and at proper temperatures. For example, the nutritive value of wheat can be improved by cooking at 100°C. At this temperature, starch granules form soluble and digestible compounds. Nutritive value, however, decreases with overheating (Lang, 1970). This improved digestibility of protein and starch with heating may explain the good growth of chicks fed Treatment 15. Nutritive value was lost due to the Maillard reaction, but this was offset in chicks fed Treatment 15, who had better weight gains and feed intakes, possibly due to moderate heating which increased protein and starch digestibility. This may have counteracted the adverse effects of the Maillard reactants. However, at the 20-minute-browning level, nutritive values in digestibility were lost through the binding of amino acids in Maillard products.

Determination of Fiber and Nitrogen in Feeds and Chick Feces

Analysis of variance and the Duncan Multiple Range Tests were used to test if there were significant

differences among the percentages of fiber and nitrogen in the feeds and in the chick feces for different treatments.

Determination of Fiber and Nitrogen in the Feeds

Determination of Neutral-Detergent Fiber (NDF) in the Feeds.

The NDF percentages, determined both with and without added sodium sulfite, in the feeds for each treatment is given in Table XVI and Table XVII. Both tables show that by either method the percentage of NDF increased as the browning levels of the treatment increased. The percentage of NDF in Treatment 20 was higher than the percentage of NDF in the other treatments. A similar result was found by Van Soest (1965) which showed that a fresh orchard-ladino forage mixture given different heating treatments increased apparent dietary fiber content as the heating time or temperatures increased. Comparing both methods, the percentage of NDF determined with added sodium sulfite for each treatment was smaller than the percentage of NDF determined without added sodium sulfite. Sodium sulfite is used in the NDF procedures in order to attack disulfide bridges and the linkages between the aromatic compounds in the protein chains. Sodium sulfite, however, may cause the loss of lignin subunits and also attack the Maillard polymers which have the effect of lowering the NDF determination (James and Theander, 1981). Statistical analysis showed that there was no significant

TABLE XVI

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF NDF PERCENTAGE DETERMINED WITH ADDED SODIUM SULFITE IN THE FEEDS (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping		
20	2	1.2806	A		
15	2	0.7215		B	
10	2	0.6735		B	
5	2	0.6271		B	C
$II_1(0)$	4	0.5114			C

* Means with the same letter are not significantly different.

TABLE XVII

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF NDF PERCENTAGE DETERMINED WITHOUT ADDED SODIUM SULFITE IN THE FEEDS ($\alpha = 0.05$)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	2	1.2988	A
15	2	0.8714	B
10	2	0.6736	C
$II_1(0)$	4	0.5879	C C C C
5	2	0.5798	C

* Means with the same letter are not significantly different.

differences between the two methods used to determine thepercentage of NDF (Table XXX. Appendix). However, since the use of the sodium sulfite caused a loss (although not significant) in apparent fiber, it is recommended that this reagent not be used.

Results of Neutral-Detergent Fiber (NDF) with Added Sodium Sulfite. Statistical analysis showed that there were significant differences among the percentages of NDA determined with added sodium sulfite for different treatments (Table XXXI. Appendix). Table XVI shows that the percentage of NDF determined with added sodium sulfite for Treatment 20 was significantly higher than the percentages of NDF for the other treatments. There were no significant differences between the percentages of NDF for Treatments $II_1(0)$ and 5. The percentages of NDF for Treatments $II_1(0)$ and 5 were lower than the percentages of NDF for Treatments 10, 15, and 20.

Results of Neutral-Detergent Fiber (NDF) Tests of Feeds Without Added Sodium Sulfite. Statistical analysis showed that there were significant differences among the percentages of NDF for different treatments (Table XXXI. Appendix). Table XVII shows that the percentages of NDF for Treatment 20 was significantly higher than the percentages of NDF for the other treatments. The percentage of NDF for Treatment 15 was significantly different from the percentages of NDF for the other treatments. There were no significant differences among the percentages of NDF for Treatments $II_1(0)$, 5, and 10. The percentage of NDF for Treatments $II_1(0)$, 5, and 10

were significantly lower than the percentages of NDF for Treatments 15 and 20.

Determination of Acid-Detergent Fiber (ADF) and Lignin in the Feeds

The percentages of ADF for each treatment is given in Table XVIII. Although the percentage of ADF for each treatment did not increase consistently as the browning levels of the treatments increased, the percentage of ADF for Treatment 20 was higher than the percentages of ADF for the other treatments. Statistical analysis showed that there were signifcant differences among the percentages of ADF for different treatments (Table XXXI. Appendix). Table XVIII shows that the percentage of ADF for Treatment 20 was significantly higher than the percentage of ADF for the other treatments. However, there were no significant differences among the percentages of ADF for Treatments $II_1(0)$, 5, 10, and 15.

The percentage of lignin for each treatment is shown in Table XIX. It appeared that the percentage of lignin for each treatment increased as the browning levels of the treatments increased. Statistical analysis showed that there were significant differences among the percentages of lignin for different treatments (Table XXXI. Appendix). Table XIX shows that the percentage of lignin for Treatment 20 was significantly higher than the percentages of lignin for the

TABLE XVIII

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF ADF PERCENTAGE IN THE FEEDS (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	2	0.64615	A
10	2	0.37620	B
15	2	0.37420	B
II_1(0)	4	0.34202	B
5	2	0.34150	B

* Means with same letter are not significantly different.

TABLE XIX

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF LIGNIN PERCENTAGE IN THE FEEDS (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	2	0.20360	A
15	2	0.09222	B
10	2	0.07687	B B B
5	2	0.06454	B B
$II_1(0)$	4	0.05053	B B

* Means with the same letter are not significantly different.

other Treatments $II_1(0)$, 5, 10, and 15. However, there were no significant differences among the percentages of lignin for Treatments $II_1(0)$, 5, 10, and 15.

Determination of Nitrogen in the Feeds

The results (Table XX) showed that nitrogen percentages in the feeds decreased when the browning levels of the treatments increased. It is not known why the nitrogen levels decreased with browning. Although amino acids may be bound during browning, the nitrogen determination method used would reflect the presence of both bound and unbound nitrogen groups. It is possible that some nitrogenous material was lost during browning. Statistical analysis showed that there were significant differences among the percentages of nitrogen in different treatments (Table XXXI. Appendix). Table XX shows that there were no significant differences among the percentages of nitrogen in Treatments $II_1(0)$, 5, and 15. There were no significant differences between the percentages of nitrogen in Treatments 10 and 20. The percentages of nitrogen in Treatments $II_1(0)$, 5, and 15 were higher than the percentages of nitrogen in Treatments 10 and 20.

Determination of Fiber and Nitrogen in the Chick Feces

The feces were darker brown in color and lower in

TABLE XX

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF NITROGEN PERCENTAGE IN THE FEEDS (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping		
II_1(0)	4	2.6401	A		
5	2	2.4606	A A	B	
15	2	2.4145	A A	B B	
10	2	2.3501		B B	C
20	2	2.1548			C C

* Means with the same letter are not significantly different.

moisture content as the browning levels of the feeds increased.

According to the literature, the dietary fiber content of food can be increased by Maillard compounds, which have the physical and chemical properties of lignin (Van Soest, 1965, 1978 and 1982; Van Soest and McQueen, 1973; Cummings, 1978). The results in this study showed that the percentages of NDF and lignin in the feeds increased with browning. It is possible that characteristics of dietary fiber, such as water-holding capacity and bulk density, may influence the bulk and water content in the feces, but this was not established in this study.

The results (Table XXI) show that the moisture percentage in the feces of chicks fed the browned Treatments 5, 10, and 20 was not higher than chicks fed the unbrowned treatments or the browned Treatment 15. The ratio of feed intake to dry weight of feces did not increase consistently as browning levels of the treatments increased. This study, therefore, does not show that increasing the percentages of NDF and lignin in the feeds with browning affects the moisture percentage and dry weight of feces.

Determination of Neutral-Detergent Fiber (NDF) in the Chick Feces

The percentage of NDF determined with added sodium sulfite or without added sodium sulfite in the chick feces from each treatment is given in Table XXII and Table XXIII.

TABLE XXI

THE EFFECT OF DIFFERENT TREATMENTS ON FEED INTAKE, DRY WEIGHT AND MOISTURE PERCENTAGE OF CHICK FECES, AND RATIO OF FEED INTAKE TO DRY WEIGHT OF FECES

Treatment	Average daily feed intake per chick (gms)	Average dry weight of feces per chick (gms)	Ratio of feed intake to dry weight of feces	% Moisture content of feces per chick
II_2	39.6	13.8	2.9:1	67.4
I	39.4	12.1	3.3:1	66.8
$II_1(0)$	37.4	11.1	3.4:1	64.4
5	36.2	17.9	2.0:1	54.9
10	30.1	11.0	2.7:1	45.8
15	35.8	12.2	2.9:1	64.0
20	21.5	6.3	3.4:1	58.1

TABLE XXII

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF NDF PERCENTAGE DETERMINED WITH ADDED SODIUM SULFITE IN CHICK FECES (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	8	4.5305	A
15	8	2.4035	B
10	6	2.0771	B C
II_2	4	1.6440	B C D
$II_1(0)$	12	1.6067	C D
5	4	1.4076	C D
I	12	1.2068	D

* Means with the same letter are not significantly different.

TABLE XXIII

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF NDF PERCENTAGE DETERMINED WITHOUT ADDED SODIUM SULFITE IN CHICK FECES (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	8	5.5523	A
15	8	2.6695	B
10	6	2.4272	B
$II_1(0)$	12	2.0195	B
II_2	4	1.8181	B
5	4	1.5227	C
I	12	1.3865	D

* Means with the same letter are not significantly different.

Comparing both methods, the percentage of NDF in the chick feces determined with added sodium sulfite from each treatment was less than that determined without added sodium sulfite. The reason has been mentioned previously in the evaluation of NDF in the feeds. Statistical analysis showed that there were significant differences between the two methods of determining NDF percentage in the chick feces from different treatments (Table XXX. Appendix). As in the NDF analysis of the feeds, when sodium sulfite was used as one of the reagents, there appeared to be a loss of fiber. Therefore, it would appear that it would be better not to use sodium sulfite.

Results of Neutral-Detergent Fiber (NDF) Tests in the Chick Feces with Added Sodium Sulfite. The results (Table XXII) show that the percentages of NDF in the feces of chicks fed the different treatments increased as the browning levels of the treatments increased, with the exception of Treatment 5. Statistical analysis showed that there were significant differences among the percentages of NDF in the feces from different treatments (Table XXXII. Appendix). Table XXII showed that the percentage of NDF in the feces from Treatment 20 was significantly higher than the percentages of NDF in the feces from the other treatments.

Results of Neutral-Detergent Fiber (NDF) Tests in the Chick Feces Without Added Sodium Sulfite. Statistical analysis showed that there were significant differences among the percentages of NDF in the feces from different treatments

(Table XXXII, Appendix). Table XXIII also shows that the percentage of NDF in the feces from Treatment 20 was significantly higher than the percentages of NDF in the feces from the other treatments.

Determination of Acid-Detergent Fiber (ADF) and Lignin in the Chick Feces

Determination of acid-detergent fiber (ADF) in the chick feces was made following the AOAC approved method without α-amylase pretreatment. The results (Table XXIV) show that the percentage of ADF in the feces from each treatment was even higher than the percentage of NDF determined by the two methods. It might be that the percentage of ADF determined without giving α-amylase pretreatment contained some residues of starch gel which would increase the percentage of ADF. Although the percentages of ADF in the feces from different treatments did not increase consistently as the browning levels of the treatments increased, the percentage of ADF in the feces from Treatment 20 was still higher than the percentages of ADF in the feces from the other treatments.

Statistical analysis showed that there were significant differences among the percentages of ADF in the feces from different treatments (Table XXXII. Appendix). Table XXIV shows that the percentage of ADF in the feces for Treatment 20 was significantly higher than the percentages of ADF in

TABLE XXIV

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF ADF PERCENTAGE IN CHICK FECES (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	8	3.4365	A
$II_1(0)$	14	2.8338	B
15	8	2.7835	B
II_2	4	2.5757	B
10	6	2.5354	B
I	12	1.6162	C
5	4	1.4454	C

* Means with the same letter are not significantly different.

the feces from the other treatments. There were no significant differences among the percentages of ADF in the feces from Treatments II_2, $II_1(0)$, 10, and 15. There were also no significant differences between the percentages of ADF in the feces from Treatments I and 5. However, the percentages of ADF in the feces from Treatments I and 5 were significantly lower than the percentages of ADF in the feces from the other treatments.

The percentage of lignin in the chick feces from each treatment is given in Table XXV. The percentage of lignin in the feces from each treatment did not increase consistently as the browning levels of the treatments increased. The determination of lignin percentage might have been affected due to ADF being determined without giving α-amylase pretreatment. Statistical analysis showed that there were significant differences among the percentages of lignin in the feces from different treatments (Table XXXII. Appendix). Table XXV shows that the percentage of lignin in the feces from Treatment 20 was significantly higher than the percentage of lignin in the feces from the other treatments. There were no significant differences between the percentages of lignin in the feces from Treatment I and 5. However, the percentages of lignin in the feces from Treatment I and 5 were significantly lower than the percentages of lignin in the feces from the other treatments.

TABLE XXV

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF LIGNIN PERCENTAGE IN CHICK FECES (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	8	0.79511	A
II_2	4	0.57355	B
$II_1(0)$	14	0.56819	B
10	6	0.56135	B
15	8	0.48159	B C
I	12	0.38988	C D
5	4	0.29707	D

* Means with the same letter are not significantly different.

Determination of Nitrogen in the Chick Feces

Maillard complexes such as bound amino acids are digestive-enzyme resistant and increase nitrogen excretion in the feces and urine (Porter and Rolls, 1971; Knipfel, 1981; Frazier et al., 1953).

Knipfel (1981) studied the effect of casein combined with different kinds of carbohydrates such a glucose, fructose, sucrose, etc., autoclaved at 121°C (249.8°F) for different periods of time on rats. The results showed that the casein-glucose or fructose mixture that was heated for 20 minutes and fed to the rats resulted in increased nitrogen content in the feces. When autoclaved for 140 minutes, the casein-sucrose mixture fed to the rats caused increased fecal nitrogen excretion. The results (Table XXVI) in this study show that Treatment 20, which was heated for 20 minutes, resulted in increased fecal nitrogen excretion.

Statistical analysis showed that there were significant differences among fecal nitrogen percentages from different treatments (Table XXXII. Appendix). Table XXVI shows that fecal nitrogen percentage from Treatment 20 was significantly higher than other fecal nitrogen percentages from the other treatments. Fecal nitrogen percentage from Treatment 5 was significantly lower than other fecal nitrogen percentages from the other treatments.

TABLE XXVI

THE DUNCAN MULTIPLE RANGE TEST FOR MEAN VALUES OF NITROGEN PERCENTAGE IN CHICK FECES (α=0.05)

Treatment	No. of Analyses	Mean (%)*	Duncan Grouping
20	8	5.2447	A
$II_1(0)$	14	4.0178	B
15	8	3.8374	B C
I	12	3.6504	B C
II_2	4	3.4009	C
10	6	3.3613	C
5	4	2.7313	D

* Means with the same letter are not significantly different.

Testing the Hypotheses

The first hypothesis (H_1) stated that there would be no significant differences in chick weight gains when the bread ingredients feed were browned for II_1(0), 5, 10, 15, and 20 minutes at 350° (176°C). According to statistical analyses and the Duncan Multiple Range Tests, there were significant differences ($P<0.05$) among weight gains of chicks fed the browned Treatments II_1(0), 5, 10, 15, and 20 over different time intervals. Chicks fed the 20-minute-browned feed had significantly less growth than chicks fed the other treatments. Chicks fed Treatment 15 had the greatest growth of all the browned feeds and their gains were as good as or better than the gains associated with the unbrowned feeds (Table X, XI, XII, XIII, and XIV). The researcher failed to accept H_1.

The second hypothesis (H_2) stated that there would be no significant differences in chick weight gains when the unbrowned feeds were prepared using J. T. Baker Company U.S.P. dextrose or Cerelose dextrose. According to the Duncan Multiple Range Test, there were significant differences ($P<0.05$)in chick weight gains between Treatment I using U.S.P. dextrose and Treatment II_2 using Cerelose dextrose at interval time Day 0 to Day 3 (Table X, XI). There were no significant differences in chick weight gains between Treatment I and Treatment II_2 over the intervals of Day 0 to Day 5, Day 3 to Day 5, or Day 0 to Day 7 (Table

XII, XIII, XIV). The researcher did not accept H_2 for time period Day 0 to Day 3. However, the researcher could not reject H_2 for time periods Day 0 to Day 5, Day 3 to Day 5, or Day 0 to Day 7.

The third hypothesis (H_3) stated that there would be no significant differences in chick weight gains when the unbrowned feeds were prepared using Multifoods flour or Pillsbury bread flour. According to the Duncan Multiple Range Test, there were no significant differences in chick weight gains between Treatment II_1 using Multifoods flour and Treatment II_2 using Pillsbury bread flour over any of the time intervals (Table XI, XII, XIII, XIV). The researcher could not reject H_3.

The fourth hypothesis (H_4) stated that there would be no significant differences in fiber percentages of feeds or chick feces due to glucose source, flour source, or browning levels of the feeds. According to statistical analyses and the Duncan Multiple Range Test, there were significant differences ($P<0.05$) in the fiber percentages (NDF, ADF, lignin) of feeds caused by the browning levels of the feeds (Table XVI, XVII, XVIII, XIX). The fiber percentage (NDF, ADF, lignin) for Treatment 20 was significantly higher than other fiber percentages for the other treatments. The researcher could not accept H_4 concerning fiber percentages of feeds due to browning level of the feeds.

According to the Duncan Multiple Range Test, there were no significant differences in NDF percentages of chick

feces, determined by two methods, caused by glucose source, or flour source of the feeds (Table XXII, XXIII). Also, there were no significant differences in the percentages of ADF and lignin of chick feces due to flour source of the feeds, but there were significant differences ($P<0.05$) in the percentages of ADF and lignin of chick feces caused by the glucose source in the feeds (Table XXIV, XXV). The researcher could not reject H_4 concerning NDF percentages of chick feces determined by both methods, caused by glucose, or flour source of the feeds. The researcher could not reject H_4 in the percentages of ADF and lignin of chick feces due to flour source of the feed, but the researcher failed to accept H_4 concerning the percentages of ADF and lignin of chick feces caused by glucose source of the feeds.

There were significant differences ($P<0.05$) among the fiber percentages (NDF, ADF, lignin) in the feces due to the browning levels of the feeds (Table XXII, XXIII, XXIV, XXV). The percentage of fiber (NDF, ADF, lignin) in the feces from Treatment 20 was significantly higher than other percentages of fiber in the feces from the other treatments. The researcher failed to accept H_4 concerning fiber percentages in the feces caused by the browning levels of the feeds.

The fifth hypothesis (H_5) stated that there would be no significant differences in nitrogen percentage of feeds or chick feces caused by glucose source, flour source, or the browning levels of the feeds. According to statistical analyses and the Duncan Multiple Range Test, there were

significant differences ($P<0.05$) in nitrogen percentage of feeds caused by the browning levels of the feeds (Table XX). The researcher failed to accept H_5 concerning the percentage of nitrogen of feeds caused by the browning levels of the feeds.

There were no significant differences in the percentage of nitrogen of feces caused by glucose source of the feeds. However, there were significant differences ($P<0.05$) in the nitrogen percentages of feces caused by the flour source as the feces of chicks fed Treatment II_2 using the Pillsbury flour contained a significantly lower percentage of nitrogen than chicks fed Treatment $II_1(0)$ using Multifoods flour (Table XXVI). The researcher could not reject H_5 concerning nitrogen percentage of feces caused by glucose source of the feeds, but the researcher failed to accept H_5 concerning nitrogen percentage of feces caused by flour source of the feeds.

There were significant differences ($P<0.05$) in the nitrogen percentage of feces caused by the browning levels of the feeds. The percentage of fecal nitrogen from Treatment 20 was significantly higher than the percentages of fecal nitrogen from the other treatments. The percentage of fecal nitrogen from Treatment 5 was significantly lower than the percentages of fecal nitrogen from the other treatments (Table XXVI). The researcher failed to accept H_5 concerning the percentages of fecal nitrogen caused by the browning levels of the feeds.

CHAPTER V

SUMMARY, CONCLUSIONS, AND RECOMMENDATIONS

A review of pertinent literature indicates that heating of food is important for flavor, color, palatability, and appetizing aroma. However, during heating the reducing sugars in food may combine with essential amino acids, such as lysine, in the Maillard reaction forming brown, indigestible, and nutritionally unavailable products. Some researchers have stated that the Maillard polymers fed to experimental animals cause a decrease in the animal's food intake and an increase in nitrogen excretion in the feces and urine. Therefore, the Maillard reaction decreases nutritive value and protein efficiency. On the other hand, the Maillard compounds appear to increase the dietary fiber content of food which may be of positive importance in nutrition.

Knight and Hanson (1982, 1983) have completed two trials investigating the effect of the Maillard reaction on chick growth. In the first trial chicks fed rations browned for 20 or 30 minutes experienced little or no growth. In the second trial, using different sources of glucose and flour, the growth of chicks was less for the 20-minute browning level than for the shorter browning levels, but was not completely inhibited as in Trial 1. The chicks fed the

15-minute-browned feed had the best growth of all treatments These results needed to be further studied.

Summary

The purposes of this study were to test the effect of browning of feed on chick growth and to determine whether using two different sources of flour and glucose in an unbrowned feed affected the chick growth differently. This study had two parts. In the first part, five chick feeds, Treatments 0, 5, 10, 15, and 20, were prepared using Cerelose sugar and Multifoods flour, as used in the second trial by Knight and Hanson (1983). These feeds were browned for 0, 5, 10, 15, and 20 minutes at 350^{o} (176.7^{o}). In the second part, there were three unbrowned chick feeds. Treatment I was made from U.S.P. dextrose and Pillsbury bread four as used in the first trial by Knight and Hanson (1982). Treatment II_2 used Cerelose sugar as used in the second trial and Pillsbury bread flour as used in the first trial. Treatment II_1 which had the same composition, i.e., Cerelose sugar and Multifoods flour, as used in the browned feeds, was compared with Treatment I and Treatment II_2.

The effect of all eight treatments on the growth rate of chicks, fiber (NDF, ADF, lignin), and nitrogen percentages in the feeds and chick feces were determined objectively.

Chick weight gains were analyzed statisticaly by using analysis of covariance. Feed intake of chicks, fiber

(NDF, ADF, lignin), and nitrogen percentages in the feeds and chick feces were analyzed by the analysis of variance (ANOVA). The Duncan Multiple Range Test was used to test the significance of differences among means for the data at the 0.05 level of significance.

Conclusions

Analyses of the data generated in this research project showed the following results and led to certain inferences.

Chick Weight Gains

Statistical analyses showed that there were significant differences ($P<0.05$) in weight gains among chicks when the feed had been browned for 0, 5, 10, 15, and 20 minutes at 350°F (176°C) (Table X, XI, XII, XIII, and XIV). The chicks fed the 20-minute-browned feed had significantly less weight gain than the chicks fed the other treatments. The chicks fed Treatment 15 had the greatest growth of all the browned feeds and their gains were as good as or better than the gains associated with the unbrowned feeds. This finding would indicate that a moderate browning (the 15-minute level in this study) was beneficial, but browning for either more or less time caused decreased gains.

Although there were significant differences in chick weight gains between Treatment I using U.S.P. Dextrose and Treatment II_2 using Cerelose dextrose at time interval Day 0

to Day 3 (Table X, XI), there were no significant differences in chick weight gains between Treatment I and Treatment II_2 over the intervals of Day 0 to Day 5, Day 3 to Day 5, or Day 0 to Day 7 (Table XII, XIII, XIV). There were also no significant differences in chick weight gains between Treatment II_1 using Multifoods flour and Treatment II_2 using Pillsbury bread flour over any of the time intervals (Table XI, XII, XIII, XIV). It was concluded that substituting either of the brands of glucose or flour in the unbrowned chick feeds did not affect the weight gains of the chicks.

Fiber in Browned Feeds and Chick Feces

There were significant differences ($P<0.05$) in the percentage of fiber (NDF, ADF, lignin) of feeds caused by the browning levels of the feeds. The results showed that the percentages of NDF determined by two methods and lignin in the feeds tended to increase as the browning level of the treatment increased. The percentage of ADF in the feeds did not increase consistently with browning, but all fiber components were significantly higher for Treatment 20 than for the other treatments (Table XVI, XVII, XVIII, XIX).

In the feces associated with treatments with browned feeds, there were significant differences ($P<0.05$) among the fiber percentages (NDF, ADF, lignin) in the feces from different treatments. The results showed that the percentages of NDF determined by both methods in the feces increased as

the browning levels of the treatments increased, with the exception of Treatment 5 (Table XXII, XXIII). Although the percentage of the ADF and lignin in the feces associated with the browned treatments did not increase consistently as the browning levels of the treatments increased, the percentages of ADF and lignin in the feces from Treatment 20 were higher than in the feces from the other treatments (Table XXIV, XXV).

Fecal Fiber Associated With Unbrowned Feeds

In the treatments involving the unbrowned feeds, there were no significant differences in the percentages of NDF of feces, determined by two methods, caused by glucose source, or flour source in the feeds (Table XXII, XXIII). Also there were no significant differences in the percentages of ADF and lignin of feces due to flour source of the feeds, but there were significant differences ($P<0.05$) in the percentages of ADF and lignin of feces caused by the glucose source in the feeds, with the feces of chicks fed the U.S.P. dextrose feed showing less of those two fibers (Table XXIV, XXV).

Nitrogen in the Feeds and Chick Feces

Statistical analysis showed that there were significant differences ($P<0.05$) in nitrogen percentage of feeds caused

by the browning levels of the feeds. The results showed that the percentage of nitrogen in the feeds decreased as the browning levels of the treatments increased (Table XX). Since the Kjeldahl test indicates total nitrogen irrespective of how it was combined, this finding would imply that nitrogenous products were lost during the heating required for browning.

Statistical analysis showed that there were no significant differences in the nitrogen percentage of feces caused by glucose source of the feeds. However, there were significant differences ($P<0.05$) in the nitrogen percentage of feces caused by the flour source, as the feces of chicks fed Treatment II_2 using the Pillsbury flour feed contained a significantly lower percentage of nitrogen than Treatment $II_1(0)$ using Multifoods flour (Table XXVI). There were also significant differences ($P<0.05$) in the nitrogen percentage of feces caused by the browning levels of the feeds; the percentage of fecal nitrogen excretion was highest in chicks fed Treatment 20, and lowest in chicks fed Treatment 5 (Table XXVI). This could be reflecting extra nitrogen products being excreted through the kidney as a result of non-limiting amino acids in the diet being used as an energy source.

Recommendations

Recommendations are as follows:

1) In this study the chicks were fed the assigned

rations for seven days. Studies of chicks fed these rations for two weeks or longer should be performed to determine if chick growth, organ development, or fiber and nitrogen content of feeds and feces would be significantly affected.

2) A comparison of growth of chicks fed browned feeds with chicks fed a regular chick ration should be done to determine whether the flour mixture is adequate for chick growth.

3) In this study the heating temperature was held constant, but the time of heating was varied. A study should be done wherein the time is held constant but the temperatures varied.

4) A variety of sugar types such as fructose, lactose, sucrose, corn sweeteners, and others should be used in the Maillard reaction to test their effects on protein binding, and chick growth.

5) A comparison of the growth of chicks fed crusts of bread baked from the experimental rations with a feed from the internal crumb should be made to see if there is more binding of amino aids in the crust.

6) In this study, the McQueen and Nicholson procedure (bacterial α-amylase) was used as a pretreatment to remove starch before determining fiber content. A comparison of different starch pretreatment procedures should be performed to see which procedure, if any, as pretreatment is better in determining the true fiber content.

7) Fat-extraction should be considered as a fiber analysis pretreatment to aid in sample filtration and accuracy of results.

8) A correlation of chick growth data obtained in the feeding trials with microbiological or chemical amino acid assays of the rations should be done to see if these more rapid means can also be used to accurately determine heat damage of protein.

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TABLE XXVII

F-VALUE TEST OF FEED INTAKE OF CHICKS
GIVEN DIFFERENT TREATMENTS
(α=0.05)

F-Value	Degree of freedom	Level of probability
7.09	6,16	P<0.05*

* P<0.05 means there are significant differences among feed intakes of chicks given different treatments.

TABLE XXVIII

F-VALUE TEST OF SLOPES OF CHICK WEIGHT GAINS
FOR ALL TREATMENTS FROM DAY 0 TO DAY 3
(α=0.05)

F-Value	Degree of freedom	Level of probability
2.32	6,64	P<0.05*

* P<0.05 means there are significant differences among the slopes of chick weight gains for all treatments.

TABLE XXIX

F-VALUE TEST OF SLOPES AND INTERCEPTS OF CHICK WEIGHT GAINS FOR DIFFERENT TREATMENTS OVER DIFFERENT TIME INTERVALS (α=0.05)

Chick weight gains by treatments / Time intervals	Testing slopes of chick weight gains			Testing intercepts of chick weight gains			Est. of slope value
	F-value	Degree of freedom	Level of probability	F-Value	Degree of freedom	Level of probality	
Day 0 to Day 3**	1.77	4,38	P>0.05	2.36	4,46	P>0.05	0.2900
Day 0 to Day 5	0.69	6,64	P>0.05	4.63	6,76	P<0.05*	0.1402
Day 3 to Day 5	1.19	6,64	P>0.05	3.06	6,76	P<0.05*	0.3272
Day 0 to Day 7	0.55	6,64	P>0.05	5.87	6,76	P<0.05*	0.0998

* P<0.05 means there are significant differences among treatments.

** Does not include testing for slopes and intercepts of Treatments I and 20 weight gains since they are far from Treatments II_2, II_1(0), 5, 10, and 15 and cannot be considered in the same group.

TABLE XXX

T-VALUE TEST OF NDF PERCENTAGES DETERMINED BY BOTH METHODS WITH ADDED SODIUM SULFITE AND WITHOUT ADDED SODIUM SULFITE IN THE FEEDS OR IN THE CHICK FECES (α=0.05)

NDF percentages determined by both methods	t-value	Degree of freedom	Level of probability
in the feeds	2.44	1,22	P>0.05
in the chick feces	5.44	1,106	P<0.05*

* P<0.05 means there are significant differences between two methods.

TABLE XXXI

F-VALUE TEST OF FIBER COMPONENTS AND NITROGEN PERCENTAGES IN TREATMENT FEEDS
(α=0.05)

Testing fiber and nitrogen percentages	F-value	Degree of freedom	Probability
NDF percentage determined with added sodium sulfite	60.37	4,7	P<0.05*
NDF percentage determined without added sodium sulfite	40.97	4,7	P<0.05*
ADF percentage	60.53	4,7	P<0.05*
Lignin percentage	16.78	4,7	P<0.05*
Nitrogen percentage	7.40	4,7	P<0.05*

* P<0.05 means there are significant differences among treatments.

TABLE XXXII

F-VALUE TEST OF FIBER COMPONENTS AND NITROGEN PERCENTAGESS IN CHICK FEEDS

(α=0.05)

Testing fiber and nitrogen percentages	F-value	Degree of freedom	Level of probability
NDF percentage determined with added sodium sulfite	25.64	6,37	P<0.05*
NDF percentage determined without added sodium sulfite	27.59	6,37	P<0.05*
ADF percentage	24.61	6,39	P<0.05*
Lignin percentage	21.51	6,39	P<0.05*
Nitrogen percentage	20.15	6,39	P<0.05*

* P<0.05 means there are significant differences among treatments.

TABLE XXXIII

MINERAL COMPOSITION OF FEED

		g/Kg
Calcium Carbonate	$CaCO_3$	55.94
Calcium Phosphate, tribasic		522.102
Potassium Phosphate, dibasic	K_2HPO_4	167.819
Sodium Chloride	NaCl	164.09
Magnesium Sulfate	$MgSO_4 \cdot 7H_2O$	65.263
Manganese Sulfate	$MnSO_4 \cdot H_2O$	12.12
Ferric Citrate		9.323
Zinc Carbonate		1.865
Cupric Sulfate	$CuSO_4 \cdot 5H_2O$	0.373
Boric Acid		0.168
Sodium Molybdate	$Na_2MoO_4 \cdot 2H_2O$	0.168
Potassium Iodide	KI	0.746
Cobalt Sulfate	$CoSO_4 \cdot 7H_2O$	0.019
Sodium Selenite	Na_2SeO_3	0.004

Note: Teklad Test Diet 73007

TABLE XXXIV

VITAMIN COMPOSITION OF FEED

	g/Kg
Thiamin HCl	2.2
Riboflavin	2.2
Calcium Pantothenate	6.6
Niacin	10.0
Pyridoxine HCl	2.2
Folic Acid	0.2
Biotin	0.04
Vitamin B_{12} (0.1% trituration in mannitol)	3.0
Menadione Sodium Bisulfite	10.0
Dry Vitamin A Palmitate (500,000 U/g)	4.0
Vitamin D_3 in VFT Casein (3000 U/g)	66.0
Dry Vitamin E Acetate (500 U/g)	24.0
Choline Dihydrogen Citrate	350.0
Corn Starch	519.56

Note: Teklad Test Diet 78475

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