ASSESSMENT OF FACTORS INFLUENCING BODY COMPOSITION OF BROILER CHICKENS

by

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GENERAL ABSTRACT

Seven experiments, involving 14,760 growing male and female broilers, were conducted to test the effect of genotype, dietary protein, amino acid profile and cereal, and lighting programs on body lipid content at market weight. Among three pure strains of broiler chickens obtained from a Canadian breeder (Shaver), the Cornish 2 strain grew the fastest and contained the highest body fat ($p<0.05$). The White Rock strain grew most slowly, and the Cornish 1 strain was intermediate. Among three commercial strains obtained from Shaver, all had very similar growth rates and carcass lipid content. Birds fed a high-protein (HP) diet had a significantly lower body lipid content ($p<0.001$), and a significantly ($p<0.001$) decreased energy retention as compared with those fed a low-protein (LP) diet. With the increase of body energy retention per kg $W^{0.75}$, an increasing proportion of retained energy was directed to body fat. An LP diet may reduce nitrogen excretion but it may result in a fat carcass. It was found that three LP diets with different essential amino acid profiles did not result in increased body lipid deposition, but they significantly reduced nitrogen excretion as compared with the control diet (NRC, 1994). Barley and wheat diets significantly reduced body fat deposition as compared with a corn diet without affecting final body weight. These results were supported by the observation that the higher the barley or wheat percentage in a diet, the lower the body fat content. Addition of wheat bran to a corn diet also reduced body fat deposition. Body lipid deposition and liver fatty acid synthesis rate were significantly lower in barley-fed birds than in corn-fed birds, with an intermediate level in wheat-fed birds. It can be concluded that barley or wheat in a diet can reduce liver fatty acid synthesis, resulting in lower body lipid content. Three increasing lighting programs (INC 2, INC 3 and INC 4) reduced early feed intake and body weight gain and resulted in complete or near complete compensatory growth in body size at 6 wk of age. The birds reared under INC 2, INC 3 and INC 4 contained significantly higher body water and protein contents ($p<0.05$) but lower body lipid content ($p<0.11$) than those reared under the control lighting program (16L: 8D). These results indicate that bird body lipid accretion can be effectively reduced by genetic selection, dietary (protein, amino acid, cereal) manipulation and environmental (lighting) control.

Key words: strain, protein, amino acid, wheat, barley, lighting, body composition, broiler

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ABBREVIATIONS

ACC	acetyl-CoA carboxylase
ADF	acid detergent fiber
AME	apparent metabolizable energy
C14:0, C16:0, C18:0	myristic, palmitic, stearic acid
C16:1, C18:1, C18:2	palmitoleic, oleic, linoleic acid
C20:4, EPA, DHA	Arachidonic, eicosapentaenoic, docosahexaenoic acid
CON1, CON2	control light 1 and 2 used in Exp. 8.1 and 8.2, respectively
Cys	cystine
D	dark
EAA	essential amino acids
Exp.	Experiment
FAS	fatty acid synthase
FFM	fat-free mass
FM	fat mass
HP	high-protein
INC1	increasing lighting program 1 used in Exp. 8.1
INC2, INC3 and INC4	increasing lighting program 2, 3 and 4 used in Exp. 8.2
INC-INT	increasing-intermittent lighting program in Exp. 8.1
INT	intermittent lighting program used in Exp. 8.1
IU	international units
L	light
LDL	low-density lipoprotein
LP	low-protein
ME	metabolizable energy
Met	methionine
n.a.	not available
n.s.	not significant, $p>0.05$
NDF	neutral detergent fiber
NEAA	non-essential amino acids
r	correlation coefficient
R^2	coefficient of determination, simple
SAA	sulfur amino acids
SDS	sudden death syndrome
SEM	standard error of the mean
TBW	total body water
TME	true metabolizable energy
TOBEC	total body electrical conductivity
VLDL	Very low-density lipoprotein

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CONTRIBUTIONS TO KNOWLEDGE

Effects of genotype, dietary protein, amino acids, and cereal, and lighting programs on body lipid accretion and growth performance were assessed in seven experiments with growing broiler chickens. New information was obtained as follows:

1. Three pure strains and three commercial strains from a breeding company and two commercial strains from a local hatchery were tested in terms of total body composition and growth performance of the broilers. The results increased the available data on growth and total body composition of broiler chickens.
2. A high protein diet resulted in decreased dietary energy retention as compared with a low protein diet in broilers.
3. In contrast to the conclusion of Boekholt et al. (1994), it was observed in the current study that as body energy retention increased, an increasing proportion of retained energy was directed to adipose tissue deposition.
4. A low protein diet with a different amino acid profile from that recommended by NRC (1994) was successful in maintaining growth performance and breast meat yield without increasing body lipid content.
5. It was observed that barley or wheat in a diet significantly reduced body lipid deposition in broiler chickens as compared with corn. This result was confirmed in two additional experiments.
6. Wheat bran added to a corn diet reduced body lipid deposition in broiler chickens. This new evidence supports the suggestion that routine use of high-fiber food will lead to weight loss in humans (Anderson et al., 1994).
7. Barley, wheat and wheat bran-corn diets reduced dietary energy retention efficiency by directing less energy retained to body lipid and more to body protein accretion as compared with the corn diet.
8. Liver fatty acid synthesis was indicated by the activities of fatty acid synthase and acetyl-CoA carboxylase and the incorporation of labeled water [^3H_2O] into liver fatty acids in the cereal study. A lower rate of liver fatty acid synthesis in birds fed the barley diet was probably responsible for their lower total body lipid content.

9. β-glucans (mainly in barley) and xylans (mainly in wheat) did not affect lipid metabolism and body lipid deposition.
10. Lighting pattern changes activity and feed intake in broilers. It was found that three increasing lighting programs (INC2, INC3 and INC4) reduced early feed intake and growth and successfully resulted in complete or near complete compensatory growth in the late stage of growth. The three lighting programs also successfully changed body composition, with higher body protein and water content and lower body lipid content for the birds reared under the three lighting programs as compared with those reared under the control lighting regime.
11. The three increasing lighting programs (INC2, INC3 and INC4) also reduced mortality due to Sudden Death Syndrome.
12. An EM-Scanner did not accurately estimate the body lipid content in broiler chickens.

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CHAPTER I

INTRODUCTION

With the economy of many developing countries improving and the global population increasing, there is an increasing demand for grains and animal products (especially meat, eggs and milk). The potential grain deficit facing the world especially in the densely populated Asian countries in the near future (Brown, 1995) may change future animal production systems since animals and humans are competing for grains. It is probably inevitable, to some extent, to reduce the grain used for animal products and to shift to poultry and egg products from beef, milk and pork in the future (Brown, 1995). Poultry products will be favored because of their higher feed efficiencies, less than 2 kg of grain for each kg of poultry weight gain as compared with 4 for pork, and 7 for beef. The potential grain crisis poses a severe challenge to poultry researchers and producers to produce more meat with less feed. One of the approaches is to reduce body fat deposition in chickens and improve feed utilization efficiency for meat production. In the current thesis, the term body or carcass fat is used equally to body or carcass lipids.

Poultry Meat is Lean

Poultry meat is ideally suitable for meeting demands for leaner meat by health conscious consumers. Broiler chicken carcasses contain much less fat (15% fat, on average, our unpublished data) than beef carcasses (29% fat, Jones, 1986) and pork carcasses (22% fat, Whittemore, 1998). Even if comparing highly trimmed steak or pork chops with poultry breast fillet, the poultry product is still by far the leanest (Leeson and Summers, 1997). Poultry fat also contains lower level of saturated fatty acids than beef and pork fat (on average, 26% vs. 47% & 44%, respectively) (NRC, 1994). However, consumers are still concerned about the obvious fat depots in the chicken (e.g., the subcutaneous and abdominal fat). About 120 g of visible fat in a 2-kg chicken (Nir et al., 1988) is removed and discarded by processors and consumers as waste before cooking. Nir et al. (1988) quantitatively dissected broilers from fat and lean lines and reported average fat distribution. Abdominal fat from the gizzard down to the cloaca, gizzard

fat, sartorial fat from both thighs, neck fat and mesenteric fat together contained about 20% of total body fat. The skin contained about 18% and the skeleton contained about 15%. The liver and feathers contained about 2.5%, striated muscles contained about 4.5% and the other muscles, intestines, lungs, kidneys, connective tissue, blood, glands etc. contained about 40%.

During the past 10 years, the proportion of broilers sold as whole birds has been decreasing quite rapidly. It was estimated that only 12% of broilers reared in the US were marketed as whole birds, with 60% being used for portioning and 28% for further processing (Anon, 1987). The increase of cut-up broiler meat requires good carcass appearance and composition (Leeson and Summers, 1997). Therefore, the demand for leaner broiler meat directs poultry researchers to study how to produce lean chickens.

Reduction of broiler body fat accretion can lower consumer's saturated fat consumption. There has been considerable publicity over the last decade concerning dietary consumption of saturated fat and cholesterol originating from various meats and eggs. A high intake of total fat, especially some saturated fats, can elevate blood cholesterol, especially low-density lipoprotein (LDL) cholesterol which is strongly related to a higher incidence and risk of coronary heart disease (NRC, 1989a). The Food and Nutrition Board's Committee on Diet and Health in the US recommended that fat intake should not exceed 30% of total dietary calories, that less than 10% of calories should be provided from saturated fat, and that dietary cholesterol intake should be less than 300 mg/d (NRC, 1989b).

Lower Physiological Threshold of Broiler Body Lipids

The fat content of 6-wk-old broiler chickens varies between 9 and 20% according to our data. It was reported that about 1 to 2.5% of total body weight was the fat present in the blood and other tissues which is physiologically necessary for normal body function (Yoshida and Morimoto, 1970; Evans, 1977). The remainder (i.e. over 85% of total body fat) is stored in adipose tissues (abdominal, subcutaneous, etc.). It is this part that the researchers are working on to reduce (Mallard and Douaire, 1988). However, this fat is not totally extra for the chicken as it has at least two physiological functions: (1) serves as energy stores. Fat yields more than twice as much energy than that of glycogen on a weight basis and, (2) serves as insulation against low

temperatures. Therefore, the fat stores (i.e. over 85% of total body fat) are physiologically useful and cannot be totally eliminated.

High dietary protein is known as one of the most effective body fat-reducing factors. When dietary protein content increased from 16 to 28%, the body fat content of broiler chickens at 7 wk of age was reduced from 16 to 10%. Thereafter, an increase of dietary protein content from 28 to 36% did not further change the body fat content (Jackson et al., 1982). When dietary energy level was reduced from 3600 to 2800 kcal/kg, fat content was reduced from 15 to about 10%. A further decrease in dietary energy to 2600 kcal/kg did not lower body fat content (Jackson et al., 1982). The combination of increasing dietary protein and decreasing dietary energy is more effective in reducing body fat in chickens. While dietary energy was gradually decreased to 2700 kcal/kg and protein level gradually increased to 26%, body fat in broiler chickens at 5 wk was reduced to 9% (Kirchgessner et al., 1978). Therefore, the minimum storage fat for the fast-growing broiler chicken at market age (6 wk) is estimated to be around 9%, depending on sex and strain. Stored fat beyond this threshold (9%) is probably what can be eliminated.

The Sources of Body Fat

There are two sources for body fat of broiler chickens: dietary origin and body synthesis. After dietary fat is hydrolyzed to fatty acids and glycerol in the gastrointestinal tract, it is absorbed into the blood. If the fatty acids are not used for energy sources in the muscle, the liver or other tissues, they will be used as materials to form triacylglycerols which are deposited in the adipose tissue. The other source of body fat is the internal synthesis in the body. Unlike swine and beef in which lipogenesis occurs primarily in adipose tissue, chickens synthesize fatty acids primarily in the liver. Other tissues, in particular adipose tissue, only have a limited capacity for lipogenesis (Larbier and Leclercq, 1994). When energy intake exceeds the broiler's maintenance and growth demand, the extra food energy is converted into body fat (Lin, 1981). Excessive amino acids of dietary source are first deaminated in the liver by transferring their amino group to α-ketoglutarate. The amino acid carbon skeleton then enters the citric acid cycle to produce acetyl-CoA that is the precursor for fatty acid synthesis. Excessive dietary carbohydrate will be oxidized to pyruvate, then to acetyl-CoA which is thereafter used to synthesize fatty acids. The

energy utilization efficiencies of dietary fat, carbohydrate and protein for body fat deposition are 0.90, 0.75 and 0.60 respectively if maintenance energy requirements are not considered (Larbier and Leclercq, 1994).

Chicken Body Lipid Accretion, Meat Flavour and Feed Efficiency

The growth of adipose tissue is a result of hyperplasia (increase in cell number) and hypertrophy (increase in cell size) of adipocytes. Up to about 6 wk of age, abdominal fat accumulation in broilers was due mainly to hyperplasia of adiocytes. Thereafter, hypertrophy of adipocytes accounted for increased size of adipose tissue (Cherry et al., 1984). In a review, Cartwright (1991) emphasized that excessive fat deposition was not due to adiocyte hyperplasia, but was due to hypertrophy. Adipocyte number was more highly correlated to development of body mass, while adipocyte volume was more highly correlated to fat deposition (Cartwright, 1991).

Body fat in broiler chickens has two main characteristics: (1) it is highly heritable: based on seven published papers, Leenstra (1986) concluded that the heritability of abdominal fat was between 0.38 and 0.75; (2) it is a highly variable component, even in highly selected pure strains. This suggests that broiler body fat can be effectively modified by genetic selection and by nutritional and environmental manipulation.

Broiler meat tenderness is related to the fat content in the meat (Leenstra, 1986). In broilers, muscle lipids are not so easily affected as abdominal fat and whole body lipids. Leenstra (1986) noted that the difference in abdominal fat between fat and lean lines was fourfold, the fat line birds had only 1.3 times as much lipid in the meat of the legs as that in the lean line birds. Merkley and Cartwright (1989) and Buyse et al. (1991) also demonstrated that inter- and intra-muscle fat content is little affected by β-agonist administration. Therefore, reduction of body fat content may not affect taste of broiler meat. However, Akiba (1988) reported that people inclined to prefer the meat from fat line birds to that from lean line birds.

Eating quality of meat is defined in terms of tenderness, juiciness, and flavour. Tenderness is the most important. It is recognized that fat plays a part in the eating quality of meat (Wood and Fisher, 1996). Fat affect myofibrillar toughness by providing insulation against the effects of rapid cooling on muscle fibers (Wood and Fisher, 1996). The flavour of meat during cooking

and eating arises from both water soluble and lipid soluble components in the tissues and interactions between them (Wood and Fisher, 1996). Juiciness depends on the amount of liquid released during mastication both from the food and saliva. Fat affects saliva production by introducing flavour compounds which stimulate saliva flow (Wood and Fisher, 1996). Ether-extractable lipids vary typically from below 1% in the breast to over 6% in the thigh and in dissected carcass muscle as a whole, values (3-6%) similar to those in the red meat species are found (Wood and Fisher, 1996). Considerable change in body fat concentration is not accompanied by an appreciate change in meat fat concentration (Leenstra, 1986).

Reduction of body fat will improve the efficiency of feed and dietary energy utilization. Adipose tissue contains about 80% fat, 2% protein and 18% water and lean meat contains 25% protein, 73% water, and 2% fat (Larbier and Leclercq, 1994). One gram of fat and protein contains 9.2 and 5.6 kcal energy, respectively (Znaniecka, 1967). The efficiency of dietary AME used for fat and protein deposition is on average 0.84 and 0.45 (MacLeod and Geraert, 1988; Boekholt, et al., 1994; Larbier and Leclercq, 1994). Therefore, the deposition of 1 g of adipose tissue and lean tissue in the body is calculated to require 9.01 and 3.32 kcal dietary AME, respectively. Namely, about 2.7 times as much feed energy is needed to deposit fat tissue as is required to deposit the same amount of lean meat tissue.

The reduction of body fat accretion and mortality in broiler production means producing more meat with less feed. This would be one of the important approaches to tackle with the potential grain competition between humans and animals used for meat production.

RATIONALE FOR THE STUDY

Based on the above discussion, the rationale for the current study was:

(1) More chickens are going for cut-up processing. Visible fat deposits are removed as waste. Reducing the fat content of broilers would reduce this waste.

(2) Fat deposition is energetically less efficient than is lean meat deposition.

(3) Chicken containing less fat is beneficial for the consumer health and economics.

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CHAPTER II

LITERATURE REVIEW

In this chapter, genetic, dietary and environmental influences on fat deposition and growth rate are reviewed. The effect of genetic selection, dietary manipulation (dietary protein, amino acid, fat, carbohydrate, energy, cereal type and fiber) and environmental factors (mainly light) are discussed.

GENETIC SELECTION

In the late 1970's, poultry researchers realized that the excess fatness of broiler chickens is a result of selection for fast growth rate. Thereafter, poultry breeders started genetic selection for fat and lean broiler chicken lines (Pym and Solvyns, 1979; Leclercq et al., 1980; Griffin et al., 1982) which resulted in various types of lean broiler chickens with different body composition emerging.

Abdominal fat is highly correlated with total body lipid content. Leclercq and coworkers (1980) conducted family selection against or for the abdominal fat proportion of siblings. Divergence in the proportion of abdominal fat appeared very early in the selection program. After seven generations of divergent selection, the high abdominal fat line and the low abdominal fat line had widely different degrees of fatness (35.5 vs 8.9 g abdominal fat/kg body weight for the fat and lean lines, respectively) (Leclercq, 1988). Cahaner (1988) also found that a high fat line of White Rock contained 70% more abdominal fat than its counterpart after seven cycles of divergent selection. Leenstra (1988) observed reduced abdominal fat as well after six generations of selection for low abdominal fat.

A biochemical approach has been taken based on the finding that the plasma concentration of very low-density lipoprotein (VLDL) was highly correlated ($r=0.6$-0.7) with body fatness (Griffin and Whitehead, 1982). After seven generations of divergent selection for or against plasma VLDL concentration, broilers in the lean line contained 145% less abdominal fat and 59% less total body lipid separately than the fat line (Whitehead, 1988a). As a consequence of the decrease in body fatness there was a 12.2% increase in body protein content, and a 14.8%

higher feed efficiency in the lean line as compared with the fat line. However, both lines of birds had the same body weight at 7 weeks of age (Whitehead, 1991).

Adipose tissue deposition costs 2.7 times the feed energy that lean tissue deposition costs. A reduction in body fat deposition will improve feed efficiency. It was reported that birds selected for improved feed efficiency contained less body fat and those selected for high feed intake had more body fat than the control birds when measured at the same body weight (Hood and Pym, 1982). Leenstra (1988) also reported that selection for improved feed efficiency resulted in reduced fatness.

The geneticists at the Shaver Poultry Breeding Farm Ltd, Ontario, Canada also conducted genetic selection for lower abdominal fat, wide muscular breast meat and high meat yield, together with growth rate and good reproduction traits. After selection, they have produced strains with different traits.

Genetically fat birds have a predisposition to fatness irrespective of feeding. When fed the same limited amount of feed, birds selected for leanness always exhibited faster growing rates, a lower level of fat synthesis and a higher feed efficiency than birds selected for fatness (Leclercq and Saadoun, 1982). Paired-feeding the same amount of a standard diet to fat and lean birds with the same initial body weight indicated that lean birds gained more weight but had significantly less total body fat and abdominal fat content (Whitehead, 1988b). Lean birds made better use of dietary protein and amino acids to synthesize body and feather protein (Saunderson, 1988), and exhibited a larger feather protein gain (Leclercq et al., 1994). Fat birds utilized more amino acid carbon to make fatty acids (Saunderson, 1988). Chickens selected for high feed consumption were 43% higher in their rate of incorporation of glucose into lipids and 27% higher in the activity of malate dehydrogenase than those selected for feed efficiency (Hood and Pym, 1982). Lilburn et al. (1982) reported that the fat line had a higher incorporation rate of tritium water into liver lipids than the lean line. Legrand et al. (1987) obtained similar results with [^{14}C] acetate.

It can be concluded that genetic selection is an effective and long-term method to produce lean chickens. Since body fat is highly heritable, genetic selection against abdominal fat or plasma VLDL concentration, or for high feed efficiency or lean meat yield has resulted in lean chickens. The lean line birds have a genetic predisposition to leanness and exhibit different

metabolic responses to dietary protein/amino acids and energy as compared with the fat line birds.

DIETARY MANIPULATION

There are indications that limits to selection for leanness were approached after about eight generations of genetic selection (Whitehead, 1990a). In addition, genetic selection was conducted in a uniform environment, and with a low-fat diet (Whitehead and Griffin, 1984). When nutrition and environment change, the body composition will be changed accordingly. For example, a high-protein diet can significantly reduce the body lipid content of both fat and lean lines (Touchburn et al., 1981; Whitehead, 1990b). These reports indicate the importance of nutritional and environmental manipulation of body composition besides genetic selection.

There are a number of nutritional factors affecting broiler body composition. The factors related to the current project are reviewed in this Chapter.

Dietary Protein

Increasing dietary protein content at a constant dietary energy content has been reported to reduce hepatic lipogenesis (Yeh and Leveille, 1969; Rosebrough et al., 1988). This effect is independent of the decrease in dietary carbohydrate accompanying the increased protein level (Leveille et al., 1975; Tanaka et al., 1983b). Rosebrough et al. (1988) emphasized that dietary protein intake *per se* is a potent regulator of avian lipid metabolism.

Jackson et al. (1982) found that as dietary protein content increased from 16 to 36% while metabolizable energy was kept constant, carcass lipid percentage decreased from 50 to 38% and protein percentage increased from 41 to 51% on a dry matter basis. Actually, the absolute amount of protein deposited in the body and the body weight remained constant except for birds fed the diet containing 16% protein which reflected the shortage of protein relative to that required for growth. Jackson et al. (1982) have shown that feed efficiency always increased when dietary protein increased and body lipid content decreased. Protein and energy retention efficiencies declined with dietary protein level. The decline of energy retention efficiency with increased dietary protein content is probably due to the increase of heat production (MacLeod,

1991; Teeter and Wiernusz, 1994). These results strongly demonstrate that the composition of broilers may be markedly modified by dietary manipulation and that the nutritional effect on body composition is no less than that of genetic selection.

A high protein diet can also increase nitrogen excretion in the manure and environmental nitrogen pollution. Nitrogen excretion can be dramatically reduced by supplying a diet with a low level of crude protein with a balance of essential amino acids (Leeson and Summers, 1997). However, in some cases, the reduction of dietary crude protein content may reduce growth rate and increase body fat deposition of broilers (Leeson and Summers, 1997).

It can be concluded that dietary protein is a powerful nutritional factor to reduce body lipid accretion. It reduces liver fatty acid synthesis and energy retention, thus results in leaner carcasses. However, a high protein diet may cause environmental nitrogen pollution. A low protein diet supplemented with limiting amino acids is a good approach to reduce nitrogen excretion, but may be accompanied by increased body lipid deposition.

Dietary Amino Acids

Dietary amino acid content not only affects body protein deposition, weight gain, and feed efficiency, but also body composition. Normally the optimum amino acid level for optimum feed: gain ratio and maximum carcass leanness is higher than that for maximum growth (Baker, 1989; and Fisher, 1994).

Grisoni et al. (1991) supplemented a basal diet containing 0.7% lysine and 18.6% protein with graded levels of L-lysine·HCl and fed the diets to male broiler chickens during 4 to 7 wk of age. Weight gain reached a plateau when the total lysine was 0.9%, but the feed: gain ratio and abdominal fat content was gradually reduced until lysine level reached 1.0 and 1.1%, respectively. Sibbald and Wolynetz (1986) supplemented a basal diet deficient in lysine with graded L-lysine·HCl and observed that fat deposition decreased when dietary lysine increased from 0.96 to 1.01 and 1.10%.

Fisher (1994) reported that weight gain, and breast yield gradually increased and fatness and feed: gain ratio gradually decreased with increasing dietary total sulphur amino acids up to 0.84 to 0.88%.

Rangel-Lugo et al. (1994) found that body lipid content was significantly reduced as dietary threonine concentration increased over the required 8.3 g/kg diet in chicks during 0 to 14 d post-hatching. Velu et al. (1972) reported that excess isoleucine (0.9% in a diet) decreased carcass lipid without affecting body weight. Increased dietary digestible arginine tended to increase protein deposition and decrease lipid deposition, especially in a fat line of birds (Leclercq et al., 1994).

Bedford and Summers (1985) reported that when dietary ratio of essential amino acids/non-essential amino acids (EAA/NEAA) increased gradually from 35/65 to 65/35, weight gain, feed efficiency and total carcass protein reached a plateau at a ratio of 55/45 irrespective of the dietary protein contents used. The proportion and amount of carcass lipids decreased and the percentage of carcass protein increased as EAA/NEAA ratio increased from 35/65 to 65/35.

Leclercq et al. (1994) found that supplementing a low-protein (14.4% protein) and low-NEAA diet with glutamic and aspartic acids to the equivalent of a 18.8% protein diet significantly reduced the adiposity of lean line birds (but not fat line) during the period of 28 to 42 d of age.

Based on the above observations, there is a possibility that body lipid deposition in chickens fed a low-protein diet is not increased when dietary amino acid balance is improved and limiting amino acids are supplemented.

Dietary Fat, Carbohydrate and Energy

Dietary carbohydrate and fat are the primary energy sources in diets. Dietary carbohydrate increases and dietary fat decreases the rate of liver fatty acid synthesis in the chicken. Yeh and Leveille (1969) observed that hepatic lipogenesis and the activities of malic enzyme and citrate cleavage enzyme were significantly reduced by feeding chicks diets high in fat (10-20%) on an iso-energetic basis as compared with a diet with only carbohydrate as the energy source. Hillard et al. (1980) and Tanaka et al. (1983a and 1983b) confirmed that either reducing dietary carbohydrate or increasing dietary fat would depress hepatic lipogenesis in growing chicks.

Dietary fat has higher body fat and energy retention efficiencies than dietary carbohydrate. When an equal amount of true metabolizable energy from corn starch or corn oil was supplemented to a basal ration for 5-7 wk old chickens, corn oil increased carcass protein, lipids

and energy retention more than corn starch (Teeter and Wiernusz, 1994). This is consistent with the observation of Jensen et al. (1970) that metabolizable energy derived from dietary fat appeared to be more efficiently utilized for tissue energy gain than the metabolizable energy from other sources. According to Larbier and Leclercq (1994), the energy efficiency for body lipid deposition is 0.85 and 0.76 for dietary fat and carbohydrate, respectively. Therefore, when dietary carbohydrate is replaced by fat on an equal energy basis, the bird should deposit more energy, lipids or protein.

Dietary energy level also significantly affects body lipid deposition. Jackson et al. (1982) reported that as dietary energy content increased from 2600 to 3600 kcal/kg by adding fat while protein level was kept constant, both relative the content and the absolute amount of body lipids gradually increased. Body protein and energy deposition efficiencies were also enhanced, thus body weight gain increased gradually with the dietary energy level, which is consistent with the results of Kirchgessner, et al. (1978). Body protein content decreased with the dietary energy level, which is due to the increase of body lipid content. The absolute amount of protein deposited in the body was the same among the diets except for the diet with 2600 kcal metabolizable energy/kg, which was low, reflecting the shortage of energy in that diet (Jackson et al., 1982).

In summary, dietary carbohydrate increases and dietary fat decreases liver fatty acid synthesis in the chicken. Dietary fat has higher utilization efficiencies for body lipid, protein and energy deposition than dietary carbohydrate. When dietary carbohydrate is replaced by fat at equal energy basis, the bird should deposit more lipids, energy or protein. Increasing dietary energy at the same protein level can significantly increase body lipid accretion in the chicken. It is, therefore, very important to formulate similar dietary carbohydrate (starch), fat/fatty acid, protein/amino acid and energy levels when studying the effect of nutritional factors on body composition.

Dietary Fatty Acid Composition

Fatty acids can be sorted into saturated (SFA), monounsaturated (one double bond, MUFA) and polyunsaturated (two or more double bonds, PUFA). The polyunsaturated fatty

acids are subdivided into two families of fatty acids, the n-6 and the n-3 fatty acids. Linoleic acid (18:2n-6) is an essential fatty acid in chickens and is the primary member of the n-6 fatty acid family and the body can convert it to the other members of its family. Linolenic acid (18:3n-3) is the other essential fatty acid in chickens (NRC, 1994) and is the primary member of the n-3 fatty acid family. These fatty acids form parts of vital body structures, perform important roles in immune system function and vision, help form cell membranes, and produce hormone-like compounds called eicosanoids (Wardlay and Insel, 1996).

Effect on fatty acid composition of body lipids

The body fatty acid composition in broilers is largely determined by the relative importance of hepatic fatty acid synthesis and exogenous dietary fat. When the diet provides large amount of fat, the body lipid composition will, to a considerable extent, be modified by the dietary fat. Olomu and Baracos (1991) found that when dietary tallow: flaxseed oil ratio changed from 6: 0 to 1.5: 4.5, the ratio of PUFA/MUFA/SFA in sartorius muscle changed from 18/43/37 to 30/37/32. Hulan et al. (1988) reported that total n-3 PUFA in edible meat lipids increased with the red fish meal or oil in a diet.

The fatty acid composition in the body is not a direct reflection of dietary fatty acid composition. Addition of saturated fat such as tallow and palm oil with high levels of SFA to a diet had less effect on fatty acid composition in chickens than addition of soybean oil (Olomu and Baracos, 1991), since saturated fatty acids undergo desaturation in the liver, e.g., stearic acid is converted to oleic acid (NRC, 1994). For the most part, the PUFA are unchanged after they are absorbed from the fowl's intestine (NRC, 1994).

Effect on body lipid deposition

Dietary fatty acid composition was reported to affect lipid deposition and metabolism in the mouse and rat. An intake of PUFA accounting for 6% of dietary energy led to a 40% reduction in liver fatty acid synthesis, but comparable levels of saturated fat were without effect when compared with a diet without addition of any fat (Clarke et al., 1977). Shimomura et al (1990) reported that rats fed a safflower oil diet accumulated less body fat than rats fed a beef tallow diet. Kitts and Jones (1996) concluded that a high PUFA/SFA ratio fat diet would result in greater fatty acid oxidation and heat production in mammals compared with a low PUFA/SFA

ratio diet. In other words, dietary PUFA after absorbed are preferentially used for oxidation to supply energy and SFA are preferentially used for storage in mammals.

The saturation of dietary fatty acids does not have significant effect on body fat deposition in broilers. Hillard et al. (1980) reported that addition of either 5 or 10% saturated (tripalmitrin) or PUFA fat (safflower oil) to a diet at the expense of equal carbohydrate energy caused a similar reduction in fatty acid synthesis and fatty acid synthase activity. Pinchasov and Nir (1992) observed that five different combinations of tallow and vegetable oil (mixture of safflower and soybean oil, 1:1) supplemented to five diets on iso-energetic basis did not result in significant ($p>0.05$) differences in broiler body fat content at 40 d of age. Similar results were reported by Hulan et al. (1988), and Olomu and Baracos, (1991). These results may indicate that the lipid metabolism of birds is, to some extent, different from mammals.

Effect on stability of body lipids

With the increase of PUFA in chicken muscle lipids, a new problem--lipid oxidation comes up. Subcellular membranes (mitochondrial, microsomal) contain phospholipids which are rich in PUFA. In addition, they are in contact with a cytoplasmic fluid containing pro-oxidants such as oxygen, transition metals, peroxidase, hydrogen peroxide and the superoxide anion radicals. Hence these membranes are very susceptible to peroxidation and are investigated to be the sites where oxidative changes are initiated in raw meat (Asghar et al., 1990).

Lipid oxidation may result in the production of undesirable odours and flavours and shorten shelf life of broiler meat. A number of factors promote lipid oxidation in meat, including α-tocopherol deficiency, selenium deficiency and high concentrations of PUFA (Lin et al., 1989). Supplementation of both α-tocopherol and BHT/BHA (two types of antioxidants) in diets significantly improves the oxidative stability of meat during refrigerated and frozen storage. In pigs, similar results have been reported (Monahan et al., 1992). Addition of saturate fat in the feed will reduce the risk of lipid oxidation in the meat. The study of Asghar et al. (1990) indicated that the rate of NADPH-induced lipid peroxidation in microsomes and mitochondria in chicken meat (breast and thigh) was dependent primarily on membrane lipid fatty acid unsaturation, and to a lesser extent, on the α-tocopherol content.

Briefly, the utilization of dietary fat, particularly vegetable oils with high PUFA, may result in a fatty acid composition in the body similar to that of the diet. Dietary fatty acid

composition does not have significant effect on body fat deposition in broilers. Lipid oxidation in the meat is associated with increased PUFA in broiler meat. Supplementation of antioxidants in oil fed to chickens can alleviate lipid oxidation occurring in meat. The lipid oxidation in meat depends primarily on the PUFA content, and to a lesser extent, on the concentration of antioxidants.

Dietary Fiber and Cereal

Dietary fiber is defined as the endogenous components of plant materials in the diet, which are resistant to digestion by endogenous enzymes produced by humans (Horn, 1997). Dietary fiber mainly includes cellulose, hemicellulose, lignins, pectins, gums and mucilages and other non-starch polysaccarides (Horn, 1997).

Numerous studies have shown that soluble fiber sources, such as oat bran, wheat germ, legume seeds, pectins, guar gum, and psyllium lower blood total and LDL cholesterol in humans and animals (Cara et al., 1992; Kritchevsky & Story, 1993; Anderson et al., 1994; Lairon, 1996). Conversely, wheat fiber such as wheat bran, representing insoluble fiber sources, have variable effects on blood cholesterol while exhibiting more marked effects on plasma triacylglycerol and triacylglycerol-rich lipoproteins in laboratory animals and humans (Cara et al., 1992; Kritchevsky & Story, 1993; Lairon, 1996).

Addition of 10 g oat bran or wheat bran significantly lowered the post-meal serum triacylglycerol increase in humans but rice bran, pea fiber and soybean fiber did not (Cara et al 1992). Decreases in postprandial serum triacylglycerols have also been observed in pigs with test meals supplemented with sugar-beet fiber, wheat bran and guar gums (Lairon, 1996). Lower fasting blood triacylglycerols have been frequently reported when high-fiber diets are fed to obese or type II diabetic hypertriglyceridemic subjects (Lairon, 1996). The reduction of serum triacylglycerol concentration in subjects consuming a meal supplemented with oat bran, wheat fiber or wheat germ was probably due to VLDL triacylglycerol reduction rather than chylomicron triacylglycerol reduction (Cara et al., 1992). In other words, the fiber sources might have affected liver fatty acid synthesis.

When corn and wheat in various proportions were fed to laying hens, the percent lipid and total lipid per liver increased as the proportion of corn increased. The total liver lipids

accumulated were the highest for hens fed grain corn and the lowest for those fed barley, or oats. Intermediate levels of liver lipids were obtained with wheat diets on iso-energetic basis (Jensen et al., 1976). Maurice and Jensen (1977) observed higher liver and plasma lipids in corn fed Japanese quail than in the wheat fed quail. These results indicate that wheat or wheat bran, barley and oats can significantly reduce the deposition of liver lipids and the concentration of plasma lipids as compared with corn in avian species.

Barley and wheat were also reported to reduce liver fatty acid synthesis in avian compared with corn. Maurice and Jensen (1977) reported significantly higher *in vivo* and *in vitro* incorporation rates of [1-^{14}C]-acetate into liver lipids with a corn diet than with a wheat diet in female laying quail. Lipids from livers of quail fed the corn diet contained significantly higher C14:0 (myristic), C16:0 (palmitic), C16:1 (palmitoleic) and C18:1 (oleic) fatty acids and lower C18:0 (stearic), C18:2 (linoleic) and C20:4 (arachidonic) fatty acids. Since endogenously synthesized fatty acids are mainly C16:0 and C 18:1 (Bottino et al., 1970), the results of Maurice and Jensen (1977) indicate that the corn fed birds synthesized more fatty acids than the wheat fed birds.

Maurice and Jensen (1979) conducted four experiments to study hepatic lipid synthesis in domestic fowl as influenced by dietary cereal on iso-energetic, iso-nitrogenous and equi-fat basis. In one experiment, with both male and female quail, total liver lipids, and lipid biosynthesis were significantly higher in corn-fed laying quail than in wheat-fed laying quail, but not for males. In another experiment, significantly lower liver and plasma lipid concentrations were observed 2 to 3 wk after feeding the wheat diet as compared with the corn diet, due to a significantly lower rate of fatty acid synthesis and triacylglycerol synthesis in livers from quail fed the wheat diet. In the third experiment with Japanese quail, feeding the wheat diet for 6 wk significantly reduced the rate of liver lipogenesis measured with [1-^{14}C]-acetate and liver lipid content as compared with feeding the corn diet. In the fourth experiment with female laying chickens, total liver lipids and the activity of fatty acid synthase were significantly elevated in birds fed the corn diet than in those fed the wheat diet.

These studies show that dietary fiber and cereal can affect lipid metabolism in humans and birds. Birds fed a corn diet exhibit higher liver and plasma lipid concentrations and higher rates of liver fatty acid synthesis than birds fed a wheat diet or a barley diet. Therefore, the utilization of wheat or barley in a diet to replace corn as the main energy source might lead to leaner

chicken carcasses since the liver is the major organ in poultry to synthesize fatty acids. To the author's knowledge, the literature does not contain such reports.

ENVIRONMENTAL FACTORS

A number of environmental factors affect bird performance and body composition, such as temperature (Howlider and Rose, 1987), oxygen (Teeter and Wiernusz, 1994), feeding regime, e.g., crumbled vs. mash diet (Marks and Pesti, 1984), cage vs. floor feeding (Deanton et al., 1974), lighting programs (Newcombe et al., 1992). Here just lighting programs are reviewed.

Lighting plays an important role in poultry production. Regardless of light intensity or photoperiod, light influences production mainly by changing the feeding pattern and activity of broilers. Continuous light or 23 h light: 1 h dark with low light intensity is widely used in commercial broiler production with the attempt of obtaining maximum growth, but is accompanied by a high incidence of leg abnormalities and mortality (Leeson and Summers, 1997). Intermittent lighting schemes, increasing photoperiod lighting or light intensity has been reported to reduce leg abnormalities and mortality, due to reduced early growth and increased activity of broilers (Classen and Riddell, 1989; Blair et al., 1993). Also, lighting programs have been investigated as a method to reduce broiler lipid deposition (Cave, 1981; Ketelaars et al., 1986; Newcombe et al., 1992). Convincing reports that lighting programs reduce broiler body lipid deposition are very few and the published reports are also not consistent.

Intermittent Lighting Programs

There are inconsistent results about the effect of intermittent lighting programs on body lipid deposition. Cave (1981) reported that birds reared under an intermittent lighting (1 h light and 3 h dark in every 4 h) had similar body weight, slightly better feed efficiency, less leaf fat and gizzard fat compared with those kept under constant light. Cave et al. (1985) also observed a reduction in abdominal fat content with an intermittent light program for seven of eight broiler genotypes kept under the intermittent light program. Buyse et al. (1996) found that both male and female broilers reared under 1L: 3D had a lower abdominal fat content at 28 d of age

compared with controls raised in constant light. At 41 d of age, differences in abdominal fat content remained for males but not for females.

Beane et al. (1979) observed that birds under an intermittent light program (1L: 2D) tended to have more abdominal fat, especially in females, a similar 8-wk body weight and a better feed conversion ratio compared with birds under constant light. Malone et al. (1980) reported similar results with an intermittent light program (2L: 4D) versus constant light. It seemed that feed efficiency was consistently improved under an intermittent light program regime. Ketelaars et al. (1986) attributed a 9% increase in abdominal fat in birds kept under an intermittent light program (1L:3D) to a reduction in maintenance energy requirement compared to control birds housed under constant light.

Simmons (1982) also noted a superior feed conversion and a heavier body weight for birds under intermittent light regimens (1L: 2D and 1L: 7D). Ketelaars et al (1986) pointed out that the positive effect of intermittent light regimens on feed conversion could not be explained by better digestion nor by better metabolizability of gross energy for the birds reared under the intermittent light program. As a matter of fact, it is because the birds reared under an intermittent light program are quite docile during the dark period and so expend less energy on maintenance (Leeson and Summers, 1997). Compensatory growth is probably another explanation for the better feed conversion with the birds reared under an intermittent light program.

In summary, birds kept under an intermittent light program may have a higher or lower abdominal fat content at market age and a better feed efficiency than those under constant light. Reduced maintenance requirements and growth compensation may explain the better feed efficiency. An intermittent light program regime at an early age actually imposes a mild feed restriction. Feed restriction regimens may result in lower or higher body fat content depending on the timing, severity and duration of feed restriction (Zubair and Leeson, 1996).

Increasing Lighting Programs

An increasing lighting regime is where photoperiod is increased gradually (e.g. increasing 4 h/wk) or abruptly from the shortest light hours (e.g. 6L: 18D at Day 7 post-hatching) to 23-h light at 5 wk of age. Increasing lighting programs did not reduce broiler body lipid content based

on the literature, although it did improve feed efficiency and bird health. Charles et al. (1992) observed that broiler body composition was not affected by an increasing lighting program compared with constant light. Significantly lower growth rate, feed intake and a marked higher feed efficiency were observed during the first 3 wk, thereafter growth rate was higher for birds housed under the increasing lighting program compared with birds housed under the constant light. Newcombe et al (1992) found that there was 10% more abdominal fat in birds reared under an increasing lighting program (55.4 vs. 49.4 g abdominal fat/bird) than in birds reared under constant light, although birds reared under the increasing lighting program and constant light had similar final body weight at 6 wk of age.

From above, it may be concluded that an increasing lighting regime reduces feed intake and early growth rate and improves feed efficiency before 3 wk of age. By market age, there is no significant difference in body weight, or birds reared under an increasing lighting program are slightly heavier as compared with those reared under constant light due to compensatory growth.

Although not reduced bird body lipid deposition in the published studies, increasing lighting programs still have possibility to reduce body fat accretion since the feed intake pattern associated with increasing lighting programs is similar to that of early feed restriction. Early feed restriction regimens were reported to lower body fat content (Plavnik and Hurwitz, 1991; Jones and Farrell, 1992).

REVIEW SUMMARY

Genetic selection is an effective means to produce lean chickens. However, total body composition data of modern commercial and pure strain broiler chickens are relatively limited in the published literature. Increasing dietary protein content is a powerful nutritional method to reduce body lipid deposition. However, literature reports of the effect of dietary protein level on the partition of retained energy between body lipids and protein are inconsistent in the literature. More research is needed to study the effect of dietary protein on the partition of retained energy. In addition, a high-protein diet may cause environmental nitrogen pollution. A low-protein diet with supplementation of limiting amino acids can dramatically reduce nitrogen excretion, but may be accompanied by increased body lipid deposition. Elevating dietary limiting amino acids or excess lysine, methionine and other essential amino acids not only increases body weight gain

and feed efficiency, but also reduces body lipid accretion. It was assumed that body lipid deposition of chickens fed a low-protein diet might not be increased when dietary amino acid balance is improved and limiting amino acids are supplemented. Dietary fiber and cereal can affect lipid metabolism in humans and birds. Birds fed a corn diet exhibit higher liver and plasma lipid concentrations and higher rates of liver fatty acid synthesis than those fed a wheat or a barley diet. Therefore, the utilization of wheat or barley in a diet to replace corn as the main energy source might lead to leaner chicken carcasses. Both intermittent and increasing light regimes can reduce bird feed intake at an early age, by imposing mild feed restriction. Feed restriction regimens were reported to lower body fat deposition. Therefore it is possible to devise a lighting program to reduce body lipid deposition without affecting final body weight of broilers.

OBJECTIVES OF THE STUDY

Based on the above literature review, the objectives of the current study are as follows:

1. Estimate the body composition of modern broiler chickens of different genotypes.
2. Further study the effect of dietary protein levels on performance and body composition of broiler chickens, with emphasis on the effect of a high-protein diet on energy retention and partition.
3. Study the effect of a low protein diet with different amino acid profile on body composition and nitrogen excretion.
4. Study the effect of dietary cereals on body composition and liver fatty acid synthesis.
5. Devise a lighting program that gives good productivity and reduces chicken body lipid deposition.

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CHAPTER III

EVALUATION OF PERFORMANCE AND BODY COMPOSITION OF DIFFERENT STRAINS OF MODERN BROILER CHICKENS

ABSTRACT

Two experiments were conducted to estimate performance and body composition of commercial strains and pure strains of broiler chickens. Exp. 3.1 was a 2 strain × 2 diet × 2 barrier level × 2 sex factorial arrangement. A total of 2,400 Peterson × Arbor Acre and Ross × Hy-line day-old chicks (male and female) was raised to 42 d of age. One half of 48 floor pens had a wood beam barrier each. The two strains had very similar weight gain, feed efficiency, feed intake and body composition. At 3 wk, the chick carcass contained 67.9% water, 18.2% protein, 11.3% fat, and 2.4% ash, and at 5 wk, chicken carcass contained 63.9% water, 17.8% protein, 15.9% fat and 2.2% ash across strains, sex, and diets. The barrier did not affect growth performance, or alter body composition and feed: gain ratio. Males grew faster and were leaner than females. The effect of protein level is reported in Chapter IV.

A total of 3,840 day-old chicks from six strains from Shaver Poultry Breeding Farm was tested in Exp. 3.2. It was a factorial experiment including three factors (strain, barrier, and sex). The three commercial strains showed very similar growth rate, with Starbro × Starbro slightly faster, Starbro × Hub Hi-Y slightly more slowly ($p<0.05$), and Starbro × Ross intermediate. The three strains also had similar feed: gain ratio, feed intake, and carcass fat content. Among the three pure strains, Cornish 2, the fast-growing strain, grew the fastest ($p<0.01$), had the highest feed intake, body fat content ($p<0.05$) and incidence of leg problems ($p<0.01$). White Rock grew the slowest ($p<0.01$), but used less ($p<0.05$) feed for a kg weight gain during 0-6 wk than the other two pure strains. Cornish 1 was intermediate. The barrier significantly reduced leg problems ($p<0.05$). This study increased the available data on body composition of current commercial broiler chickens and of new pure strains.

Key words: genotype, body composition, barrier, broiler chickens

INTRODUCTION

Over time, the demand for broiler meat quality and production performance is changing. Initially, growth rate of broilers was the primary characteristic of concern. Direct selection of high growth rate resulted in a shorter and shorter time required to grow a broiler chicken to 2 kg body weight (Zubair and Leeson, 1996). However, there are several indirect correlated responses with this selection: an increase in fatness, leg problems, sudden death syndrome, and a loss in reproductive ability (Mallard and Douaire, 1988). Geneticists realized the increasing fatness of broilers in the middle of 1970's. A few genetic groups in the world started to work on lean chickens during the late 1970's. From then, they selected chickens with either low abdominal fat pad (Leclercq et al., 1980), low plasma VLDL (very low-density lipoprotein) concentration (Whitehead and Griffin, 1984), or high feed efficiency (Pym and Solvyns, 1979).

After several successive generations of divergent selection, broilers of the lean line were 18% lower in body fat than the unselected control chickens, and 37% lower in body fat content than the fat line chickens (Whitehead, 1988), but the lean and the fat strains showed similar growth rate.

Geneticists in Shaver Breeding Farm Ltd., Ontario, Canada have established broiler strains with different traits. For example, the Starbro female has good and consistent chick production under a variety of conditions, while the Starbro male has high meat yield. The Starbro Package can provide high numbers of chicks and high growth rate with high meat yield (the website of Shaver Poultry Breeding Farm, 1998). Some other broiler breeders with new traits are still under selection, purifying, or testing. For instance, Cornish 1 has the traits of low abdominal fat and wide muscular breast meat. Cornish 2 has a high growth rate and a broad body. However, Cornish birds usually lay only a few small eggs with poor hatchability (North and Bell, 1990). White Rock, as a dam line, lays more eggs and can be used to cross with Cornish males to produce good traits in the synthetic line (North and Bell, 1990).

Barriers and ramps were reported to increase *pectoralis* and *femorotibialis* muscle weight at 4 wk of age (Sandusky and Heath, 1987). Therefore, in two experiments, the possible effect of barriers on body composition and performance was examined.

There is very little literature reporting the total body composition of modern broiler chickens. The objective of Exp. 3.1 was to investigate the body composition of male and female

commercial broiler chickens. The objective of Exp. 3.2 was to compare the performance and body composition of three pure and three commercial strains of broiler chickens.

MATERIALS AND METHODS

Experiment 3.1

Experiment 3.1 was a factorial design (4 factors by two levels), i.e., 2 strains × 2 diets × 2 barrier levels × 2 sexes. There was a total of 48 floor pens used. Each factor level had 24 replicate pens.

Chicken Strains, diet and barrier

2,400 chicks of two commercial strains, Peterson × Arbor Acre, and Ross × Hy-line, vaccinated against Marek's disease, were obtained at day-old from a local commercial hatchery. Each strain had 600 males and 600 females. The chicks were allocated at random by strain to the 48 floor pens, 50 males or females each.

A low-protein diet and a high-protein diet (Table 4.1 in Chapter IV), based mainly on local ingredients wheat and barley were formulated to contain similar metabolizable energy and other nutrient contents except that the protein content differed. Each diet had a starter formula for 0-3 wk, and a grower formula for 3-6 wk.

Half of the floor pens had a physical wood barrier each and the other 24 pens had no barriers. The wood beam barrier, measuring 3.8 × 8.9 × 284 cm, was placed between a tube feeder and a nipple drinker and extended across the width of experimental pens, so that the chicken had to move across the barrier to feed and drink. The barrier was set up flat at 3.8 cm high above the floor litter during the first week, and vertically at 8.9 cm high when the birds were 8 to 42 d of age. The hanging-tube feeder and the 2.4-m-long nipple drinker were separately set up at each end in a pen.

Lighting and temperature

Three pens shared two incandescent light bulbs. An 18-h light/ 6-h dark (18L: 6D) lighting regime was used for all treatments. Light intensity measured 23 cm above the floor litter

and directly below the incandescent light bulb was 18 lx for 0 to 7 d and 5 lx for 8 to 42 d. Two pens shared a Shenadoah gas brooder. The temperature under the brooders and 10 cm above the litter was 33 C during 1 to 3 d and 31 C during 3 to 6 d, and later was reduced by 2 C/wk to 22 C at 35 d of age, when the brooders were switched off. Thereafter, room temperature was kept at 22 C until 42 d of age.

Experimental procedure and data collection

Chicks were allowed *ad libitum* access to feed and water. Mortality was recorded daily and cause of death was determined by post-mortem. Individual body weight of about half of the chicks in each pen was measured manually on an electronic balance at 3 and 6 wk, respectively. Feed consumption for 0-3 and 3-6 wk respectively was recorded. Five chickens per pen at each of 3 and 5 wk of age were randomly collected as samples. All the sample birds collected were scanned live in an Electro-Magnetic Scanner to obtain total body electrical conductivity (TOBEC, see Appendix I) before they were killed by cervical dislocation. The head and neck, wings at the first joint and legs at the hock joint were removed, since the wing-tip feathers, beak and feet might not be well cooked in an autoclave oven and mixed, thus affect sub-sampling. The carcasses were weighed, bagged, and frozen at -20 C until later preparation for analyses.

Experiment 3.2

Chicken strains and experimental design

This experiment was divided into two parts due to unequal numbers of chicks in the six strains offered by a commercial supplier (Shaver Poultry Breeding Farm, Ontario, Canada). In Part A, 3 commercial strains, Starbro × Ross, Starbro × Hub Hi-Y and Starbro × Starbro, 320 males and 320 females each, were tested in 24 of 48 pens. It was a 3 strain × 2 sex × 2 barrier number factorial experiment. The two barrier numbers were either no or two barriers in a pen.

In Part B, six hundred and forty male chicks from three pure strains, i.e., Cornish 1 (low abdominal fat and high breast yield), White Rock (mother side male), and Cornish 2 (high growth rate) were examined in 24 pens. It was a 3 strain × 4 barrier number factorial arrangement. The four barrier numbers were 0, 1, 2, or 3 barriers in a pen.

Barrier and diet

The barriers were the same as used in Exp. 3.1, but the barriers were set up at 3.8, 8.9 and 12.7 cm high above the surface of the floor litter during 0 to 1, 1 to 3 and 3 to 6 wk of age, respectively. The height of the barrier in this experiment was elevated during 3 to 6 wk based on the results in Exp. 3.1. The composition of the diet used in Exp. 3.2 is listed in Table 3.1.

Table 3.1 Dietary composition and nutrient content (percentage unless stated, Exp. 3.2)

Ingredients	Starter formula (0-3 wk)	Grower formula (3-6 wk)
Soybean meal	28.6	17.8
Canola meal	--	3.0
Meat meal	5.0	5.0
Corn	34.9	34.9
Wheat	25.9	32.5
Tallow	3.8	5.0
Others[a]	1.82	1.82
Total	100	100
Determined analyses		
AME, kcal/kg[b]	3042	3108
Crude protein	22.5	18.5
Ether extract	5.8	7.7
Crude fiber	3.4	3.2
Calcium	0.94	0.73
Phosphorus	0.62	0.57

[a] Others included 0.537% limestone, 0.176% mono calcium phosphate, 0.5% vitamin premix, 0.5% mineral premix, 0.1% Coxistac, and 0.025% Zinc Bacitracin. The vitamin premix supplied per kg of diet; vitamin A 9000 IU; vitamin D_3 1500 IU, vitamin E 10 IU; vitamin K_3 0.5 mg; vitamin B_{12} 0.007 mg, thiamin 0.4 mg; riboflavin 6 mg; folic acid 1 mg, biotin 0.15 mg; niacinamide 35 mg; pyridoxine 4 mg, choline chloride 1,000 mg; and Ethoxyquin 0.125 g. The mineral premix supplied per kg diet; salt (NaCl) 2g; manganese 60 mg, copper 5 mg, zinc 50 mg, selenium 0.1 mg, and iodine 0.35 mg.

[b] Calculated apparent metabolizable energy.

Experimental procedure

All chicks were fed the same diets shown in Table 3.1. The lighting regime, room temperature control and the experimental procedure were the same as in Exp. 3.1. At each of 3 and 6 wk of age, five chickens per pen were collected as samples. These chickens were scanned and processed the same way as in Exp. 3.1. Leg abnormalities (varus, valgus, leg injured or swollen) were examined at 6 wk of age while individually being weighed.

Laboratory and Statistical Analyses

The frozen chicken carcasses were cooked in a large autoclave oven (Steel Fabricating Welding Co. Ltd., W. Dundas, Ontario) at 120 C and 230 kpa for 6 h and 14 h for 3- and 6-wk chicken carcasses, respectively. The cooked carcasses were blended in a heavy-duty blender, and freeze-dried in a freeze dryer (Virtis Consol 24). The dried carcass samples were ground in a coffee grinder. Feed samples were ground to pass through a 2-mm mesh screen (Tecator, Cycloted 1093 sample mill, Sweden). Dry matter, crude fat (extracted with petroleum ether) and ash contents of carcasses, and dry matter, crude fiber, and crude fat contents of feed samples were analyzed in the laboratory using standard proximate analysis methods (AOAC, 1984). Calcium and phosphorus contents of feed were analyzed using atomic absorption spectropotometry and colorimetry methods, respectively (AOAC, 1984). Crude protein (N × 6.25) content of feed samples was analyzed with a LECO® FP 428 Nitrogen Analyzer. Carcass protein content was calculated as (100 - water % - fat % - ash %). Carcass energy content was calculated as carcass fat, g × 9.2 kcal/g + carcass protein, g × 5.6 kcal/g (Znaniecka, 1967).

All the parameters (body weight, feed intake, feed/gain ratio, body composition, etc.) were statistically analyzed for the main effect of each factor and their interactions using SAS® (SAS Institute Inc., 1996). Any significant differences between the strains or the barrier levels were further compared using Duncan's multiple range test (SAS Institute Inc., 1996).

RESULTS

Experiment 3.1

Effect of strains

The two commercial strains, Peterson x Arbor Acre and Ross x Hy-line exhibited similar performance in respect of growth rate, feed conversion, and feed intake (Table 3.2).

Table 3.2 Effect of strain, barrier and sex on growth performance and mortality (Exp. 3.1)

	Body Weight, g		Feed: gain ratio, g/g			Feed intake, g/d/bird			Mortality
	3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-6 wk %
Strain									
Pet × AA[1]	711	2146	1.44	2.04	1.83	46	140	89	3.4[b]
Ross × Hy-line	711	2160	1.43	2.06	1.83	45	141	89	6.2[a]
									**
Barrier									
0	712	2154	1.43	2.05	1.83	46	140	89	5.6[a]
1	710	2152	1.43	2.05	1.83	45	140	89	4.1[b]
									*
Sex									
Male	739[a]	2320[a]	1.42[b]	2.01[b]	1.86[b]	44[b]	130[b]	84b	6.0[a]
Female	682[b]	1988[b]	1.44[a]	2.09[a]	1.81[a]	47[a]	151[a]	95a	3.6[b]
	***	***	*	***	**	***	***	***	**
SEM[3]	2	7	0.01	0.02	0.01	0.2	1	1	0.0
N[3]	612	574	24	24	24	24	24	24	24
Interaction[2]									
Strain × Diet	***								
Barrier × Sex		*							
Strain × Barrier									*
Strain × Sex			*						

[ab] Means with different superscripts are significantly different, * p<0.05; **p<0.01; ***p<0.001.

[1] Peterson × Arbor Acre

[2] Only significant interactions are listed. The interaction between diet and sex is reported in Chapter VI.

[3] SEM: Standard error of mean; N: number of observations.

These two commercial strains, on average for both male and female, weighed about 2,150 g at 6 wk and contained 16.0% body fat at 5 wk of age. There were significant (P<0.05) two-way interactions in 3-wk body weight between strain and diet and in 6-wk body weight between barrier and sex. On the high-protein (HP) diet, Peterson × Arbor Acre birds had higher 3-wk body weight than Ross × Hy-line birds (733 vs. 721 g), while on the low-protein (LP) diet, Ross × Hy-line birds had higher body weight than Peterson × Arbor Acre birds (701 vs. 688 g). Ross × Hy-line birds had significantly higher mortality than Peterson × Arbor Acre birds (6.2 vs. 3.4 % respectively, Table 3.2, p<0.05).

At 3 wk, chick carcasses contained 67.9±1.7% water, 18.2±0.8% protein, 11.3±1.8% fat, and 2.4±0.3% ash across strains, sexes, and diets. At 5 wk, chicken carcass contained 63.9±2.1% water, 17.8±0.8% protein, 15.9±2.4% fat and 2.2±0.15% ash across strains, sexes,

and diets. Carcass protein and ash contents were quite constant while carcass fat and water were highly variable. There was no significant difference in any body component between the two strains at both 3 and 5 wk of age (P>0.05).

Effect of barrier

The physical barrier neither affected feed intake nor changed chicken's performance (Table 3.2). At 6 wk of age, male birds with a barrier had higher body weight than male birds without a barrier (2332 vs. 2308 g), but female birds with or without a barrier had an inverse pattern in 6 wk body weight (Table 3.2, Interaction of barrier × sex, $p<0.05$). The barrier reduced mortality significantly ($p<0.01$), but with Peterson × Arbor Acre birds, the barrier did not have significant effect on mortality (2.5 vs. 1.9% for 1 and 0 barrier, respectively, Table 3.2). The barrier did not alter the body composition, but male birds with the barrier contained lower body fat content at 3 wk of age than male birds without the barrier (10.1 vs. 10.7, respectively, $p<0.05$). The effect of the barrier on chicken's behavior will be reported in a separate paper elsewhere.

Table 3.3. Effect of strain, barrier and sex on carcass composition (Exp. 3.1)

	Carcass fat %		Carcass protein, %		Carcass water, %		Carcass energy[1], kcal/kg	
	3 wk	5 wk	3 wk	5 wk	3 wk	5 wk	3 wk	5 wk
Strain								
Peterson×Arbor Acre	11.4	16.0	18.2	17.9	67.9	63.9	2094	2504
Ross × Hy-line	11.3	15.9	18.2	17.8	68.0	63.8	2087	2505
Barrier								
0	11.4	15.9	18.2	18.1	68.0	63.8	2092	2507
1	11.3	16.0	18.2	18.0	67.9	63.9	2089	2502
Sex								
Male	10.4^{b}	14.8^{b}	18.3^{a}	18.0	68.7^{a}	65.0^{a}	2015^{b}	2396^{b}
Female	12.2^{a}	17.1^{a}	18.1^{b}	18.1	67.2^{b}	62.7^{b}	2168^{a}	2613^{a}
	***	***	*		***	***	***	***
SEM[2]	0.1	0.2	0.1	0.1	0.1	0.2	13	18
N[2]	120	120	120	120	120	120	120	120
Interaction[3]								
Barrier × Sex	*							

[ab] Values with a different letter differ significantly, * $p<0.05$; ** $p<0.01$; *** $p<0.001$.

[1] Carcass energy (kcal/kg) = carcass fat, g × 9.2 + carcass protein, g × 5.6.

[2] SEM: Standard error of mean; N: number of observations.
[3] Only significant interaction is listed.

Sex differences

Male chickens showed better performance (body weight and feed efficiency, $p<0.001$), higher feed intake ($p<0.001$) and higher rate of mortality ($p<0.01$) than female chickens during 0-3, 3-6 and 0-6 wk as expected (Table 3.2). Male birds had a lower feed gain ratio than female birds during 0-3, 3-6 and 0-6 wk of age, but for Ross × Hy-line birds during 0-3 wk, there was no significant difference between male and female birds in feed/gain ratio (1.42 vs. 1.43, respectively, $p>0.05$). Male chickens contained significantly lower carcass fat and higher carcass water than their female counterparts at 5 wk (14.8 vs. 17.2% for fat, 65.0 vs. 62.7% for water, respectively). Due to the differences in body fat and water content, carcass energy content was higher for females than for males ($p<0.001$). The results showing the effect of dietary protein level are tabulated in Chapter IV.

Experiment 3.2

Effect of strain

In Part A, the three commercial strains, Starbro × Ross, Starbro × Hub Hi-Y, and Starbro × Starbro exhibited similar growth rates. The Starbro × Hub Hi-Y strain had a slightly, but significantly lower body weight at 6 wk of age than the other two strains ($p<0.05$, Tables 3.4). The three strains had similar feed intake (Table 3.4), and feed conversion (feed: gain ratio) during 0-3 and 3-6 wk of age, and contained similar amounts of body lipid at both 3 and 6 wk of age (Table 3.4).

In Part B where three pure strains were compared, Cornish 2, the high growth rate strain, grew the fastest, followed by Cornish 1. White Rock grew most slowly during 0-3, 3-6 and 0-6 wk ($p<0.01$, Table 3.5). The relative feed intake values for the three pure strains were in the same order as the body weights. The White Rock strain used less feed for a kg weight gain during 3-6 wk than the Cornish 2 strain (Table 3.5). The Cornish 2 strain, besides having the highest body weight and feed intake, also had the highest body lipid content ($p<0.05$) and the highest rate of leg problems ($p<0.001$).

Table 3.4 Effect of three commercial strains, sex and two barriers on performance, carcass fat content and leg problems (Exp. 3.2)

	Body weight, g		Feed: gain ratio, g/g			Feed intake, g/bird			Carcass fat, %		Mortality	Leg
	3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	3 wk	6 wk	%	problem[2], %
Strain												
Starbro x Ross	692	2222[ab]	1.49	2.00	1.84	961	3060	3930	10.4	15.2	5.0	8.9
Starbro x Hub Hi-Y	681	2189[b]	1.49	2.00	1.84	943	3024	3887	10.4	15.4	3.5	6.0
Starbro x Starbro	691	2252[a]	1.49	2.00	1.84	953	3099	3965	10.6	15.2	4.0	7.2
SEM[1]	4	14	0.13	0.02	0.01	7	28	31	0.1	0.2	0.1	0.2
Significance		*										
N[1]	288	284	8	8	8	8	8	8	40	38	284	284
Sex												
Male	712[a]	2406[a]	1.49	1.98	1.83	993[a]	3342[a]	4235[a]	10.3	14.2[b]	5.1	7.3
Female	662[b]	2037[b]	1.49	2.02	1.85	912[b]	2780[b]	3620[b]	10.6	16.3[a]	3.3	7.5
Significance	***	***				***	***	**		***		
Barrier												
0	692[a]	2241[a]	1.48	2.00	1.84	953	3090	3957	10.4	15.4	4.2	9.9[a]
2	683[b]	2203[b]	1.51	2.00	1.84	952	3032	3898	10.5	15.1	4.2	4.9[b]
SEM[1]	4	11	0.01	0.01	0.01	6	23	26	0.1	0.2	0.1	0.1
Significance	**	*										**
N[1]	433	426	12	12	12	12	12	12	60	57	426	426
Interactions[3]												

[abcd] Means with different superscripts are significantly different, *: $p<0.05$; ** $p<0.01$; ***: $p<0.001$.

[1] SEM: Standard error of mean; N: number of observations.

[2] Leg problem: leg abnormality rate (%).

[3] All two-way interactions (strain × sex, strain × barrier, and sex × barrier) are not significant, $p>0.05$.

Table 3.5 Effect of three male pure strains and four barriers on performance, carcass fat content and leg problems (Exp. 3.2)

	Body weight, g		Feed: gain ratio, g/g			Feed intake, g/bird			Carcass fat, %		Mortality	Leg
	3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	3 wk	6 wk	%	Problem[2], %
Strain												
Cornish 1	771[b]	2591[b]	1.52	2.02[ab]	1.87[a]	1101[b]	3678[b]	4679[b]	10.1	14.3[b]	2.1	5.0[b]
White Rock	674[c]	2169[c]	1.50	1.98[b]	1.83[b]	938[c]	2976[c]	3835[c]	10.1	14.2[b]	2.1	5.7[b]
Cornish 2	803[a]	2695[a]	1.53	2.03[a]	1.88[a]	1155[a]	3862[a]	4907[a]	10.2	15.1[a]	2.4	14.1[a]
SEM[1]	4	14	0.02	0.02	0.01	15	31	35	0.2	0.2	0.1	0.2
Significance	***	**		*	*	***	**	*		*		***
N[1]	285	284	8	8	8	8	8	8	39	38	284	284
Barrier												
0	746[b]	2500	1.53	1.96[b]	1.83[b]	1067	3467	4440	10.1	14.5	2.0	9.0
1	746[b]	2450	1.51	2.01[ab]	1.86[ab]	1051	3502	4457	10.1	14.1	2.4	8.7
2	764[a]	2495	1.51	2.03[a]	1.87[ab]	1074	3545	4519	9.9	14.2	2.6	8.6
3	745[b]	2454	1.52	2.03[a]	1.88[a]	1067	3507	4480	10.3	14.4	1.8	6.1
SEM[1]	5	16	0.02	0.02	0.01	17	36	41	0.2	0.2	0.1	0.2
Significance	*			*	*							
N[1]	214	213	6	6	6	6	6	6	29	29	213	213
Strain × barrier	***	*										*

[abcd] Means with different superscripts are significantly different, *: $p<0.05$; ** $p<0.01$; ***: $p<0.001$.

[1] SEM: Standard error of mean; N: number of observations.

[2] Leg problem: leg abnormality rate (%).

Effect of barrier

In Part A, 2-barrier chickens showed significantly lower body weight than 0-barrier chickens at both 3 and 6 wk of age, e.g., 2203 vs. 2241 g/bird at 6 wk, respectively ($p<0.05$). Also, the two-barrier group exhibited significantly lower leg problems than the 0-barrier group ($p<0.01$, Table 3.4). The barrier in Part A slightly reduced feed/gain ratio and feed intake. The 2-barrier chickens had similar body lipid contents to the 0-barrier chickens.

For the three pure strains in Part B (Table 3.5), all the barrier groups showed slightly lower body weight than the non-barrier group at 6 wk ($p>0.05$). Feed/gain ratios of the chickens reared with 2 or 3 barriers were higher than that of the 0-barrier chickens ($p<0.05$), but the barrier (1, 2 or 3 barriers) did not affect the body lipid content of broiler chickens as compared with the 0-barrier group ($p>0.05$, Table 3.5). However, the more barriers in a pen, the less the leg problems.

Sex differences

As expected, male birds had higher feed intake and body weight at both 3 and 6 wk of age than their female counterparts in Exp. 3.2 ($p<0.001$, Table 3.4). However, it was surprising that the two sexes exhibited similar feed conversion values during both 0-3 and 3-6 wk ($p>0.05$). The 3-wk carcass lipid content was also similar between sexes, although the 6-wk carcass fat content was higher for the females (16.3 vs. 14.2%, $p<0.001$). The two sexes did not show significant difference in mortality and leg problems.

DISCUSSION

Performance and Body Composition

The two strains of broilers examined, Peterson x Arbor Acre and Ross x Hy-line, exhibited similar body weight gain, feed conversion, feed intake and body composition in Exp. 3.1. Ross x Hy-line exhibited significantly higher mortality than Peterson × Arbor Acre (6.2 vs. 3.4%, respectively). The higher mortality for Ross x Hy-line birds can be a significant cost to the producer. The three commercial strains (Starbro x Ross, Starbro x Hub Hi-Y and Starbro x Starbro) from Shaver exhibited different growth rates, with Starbro x Hub Hi-Y significantly

lower in final body weight than the other two strains. Feed conversion, feed intake and carcass fat contents were similar among the three commercial strains. Starbro x Hub Hi-Y was expected to have higher carcass fat content than the other two strains, as Hub Hi-Y females were selected for high abdominal fat weight. Carcass composition analysis indicated that Starbro x Hub Hi-Y contained similar 3-wk and 6-wk carcass fat (%) to the other two. Since abdominal fat is a discrete tissue that grows more rapidly and is more easily subject to changes than the total body lipid content (Cartwright, 1991), abdominal fat difference between strains may not directly reflect carcass lipid differences. The total body composition in the current study provides the breeder with useful body composition data.

The three pure strains, selected with different criteria, showed different performance and body composition. Cornish 1 was selected for low abdominal fat and high breast yield, while Cornish 2 was selected for high growth rate. After generations of selection, Cornish 1 deposited significantly lower body lipids than Cornish 2 (14.3 vs. 15.1%, respectively). Cornish 2 grew faster than Cornish 1. At 6 wk of age, Cornish 2 and Cornish 1 male birds weighed 2,695 and 2,591 g/bird, respectively. These results strongly indicate the effectiveness of genetic selection in changing body size and body composition of broilers. Cornish 2 also exhibited significantly higher leg problems (14.1 vs. 5.0, and 5.7% for Cornish 2, Cornish 1 and White Rock, respectively, $p<0.001$). The higher leg abnormalities may be related to the higher growth rate of the Cornish 2 strain (Leeson and Summers, 1997).

The White Rock strain grew more slowly than the other two pure strains due mainly to the lower feed intake. As White Rock birds grew more slowly, the maintenance requirement might account for higher proportion of AME intake and the feed/gain ratio should be higher than for the other two pure strains. White Rock birds had a lower feed/gain ratio. This is probably because White Rock birds contained relative lower body lipid, which reversed the anticipated higher feed/gain ratios.

The effectiveness of genetic selection was also displayed by the uniformity of the birds in different strains. The coefficient of variation (C.V.) of total lipid content varies between 15 and 20% (Leenstra, 1986). In the current study, C.V. of the body fat content was 5-10% for the birds obtained directly from the breeder (Shaver Poultry Breeding Farm). These results indicate that pure strains and the first filial generation exhibits very consistent performance. The more consistent the body composition and body size, the easier the production and management.

The significant interaction between strain (Peterson × Arbor Acre and Ross Hy-line) and diet (HP and LP diets) may suggest that Ross Hy-line birds had lower protein requirements since they grew better on the LP diet than Peterson × Arbor Acre birds. Barrier affected mortality in the two strains (Peterson × Arbor Acre and Ross Hy-line) differently, indicating the difference between strains.

Effect of Barriers

The barrier treatment did not affect the final body lipid content of broiler chickens in either experiment, even when the height of the barriers was increased and the number of barriers was increased up to three in a pen in Exp. 3.2. The overall increase in feed: gain ratios for birds with barriers (Table 3.4 and 3.5) suggested that the barriers do increase bird activity and energy expenditure. Apparently the increase in energy expenditure stimulated by the barriers was not sufficient to affect body composition.

In Exp. 3.1, the single barrier did not affect any of the measured parameters except reduced mortality, suggesting that the barrier was too low. The significantly different response of male and female birds to the barrier in terms of body weight may suggest that the barrier was high enough and increased female bird activity, thus reducing female body weight, but not for male birds. In Exp. 3.2, the commercial chickens with 2 barriers had significantly lower body weight at 3 and 6 wk than chickens without a barrier ($P<0.05$). The feed: gain ratio was also slightly higher for the 2-barrier groups than for the non-barrier group. These results indicated that the physical barrier in Exp. 3.2 significantly increased commercial chicken activity, thus energy expenditure and the feed: gain ratio. However, the barrier did not change the final body weight of the pure strains. The significant strain × barrier interaction on body weight gain indicates that the barrier may be still too low to change the Cornish bird activity.

The incidence of leg problems (varus, valgus, leg injured or swollen) of the commercial strains was significantly lower for the 2-barrier group than for the non-barrier group (4.9% vs. 9.9% respectively) in Exp. 3.2. The more barriers in a pen, the lower the incidence of leg problems in the pure strains (Table 3.5). However, the effect was not significant among the number of barriers ($p>0.05$). The significant interaction ($p<0.05$) between strain and barrier indicates that the barrier may be still too low to increase the Cornish bird activity and reduce

their leg problems. Leg abnormality is a big problem with fast-growing modern broiler chickens in the poultry industry (Leeson and Summers, 1997). It seems worthy to use the simple physical barriers to reduce leg abnormalities by up to 50% at the expense of body weight decline by 20 to 40 g per bird at 6 wk of age. The reduction of leg abnormalities by the barrier is mainly through increasing bird activity. The enhanced activity probably made the legs and the joints stronger.

Sex Differences

In the two experiments, male chickens always exhibited better performance and contained lower body lipid than female chickens. Male birds also had a lower feed/gain ratio than female birds, but no sex difference in feed/gain ratio was observed in Ross × Hy-line birds during 0-3 wk. This may suggest that Ross × Hy-line birds had less difference in feed conversion between sexes (Exp. 3.1). Exp. 3.4 also indicated that the three commercial strains from Shaver did not show significant ($p>0.05$) difference in feed: gain ratio between sexes. Male birds had higher mortality than female birds ($p<0.05$, in Exp. 3.1), but in Exp. 3.2, the difference was not significant ($p>0.05$). This difference may be related to strain and experimental condition.

CONCLUSION

The two commercial stains, Peterson × Arbor Acre and Ross × Hy-Line, showed very similar performance and body composition. At 5 wk, the chicken carcass contained (mean ± standard deviation) 63.9±2.1% water, 17.8±0.8% protein, 15.9±2.4% lipids and 2.2±0.2% ash across strains, sex, and diets. The three commercial strains from a breeder (Shaver Poultry Breeding Farm) also showed similar growth performance and body composition (e.g., 15.3% body lipids) in Exp. 3.2. The Cornish 1 strain, selected for low abdominal fat and high breast meat yield, contained lower body lipids than the Cornish 2 strain, selected for growth rate. Cornish 2 which grew the fastest, deposited the most body lipid, and exhibited the highest rate of leg abnormalities among the three pure strains. White Rock birds grew the most slowly but used feed more efficiently for weight gain. The results indicate that genetic selection is a very effective tool to change body size and composition in broilers. This study increased the available data on body composition of current commercial broiler chickens.

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CHAPTER IV

LIPID, PROTEIN AND ENERGY DEPOSITION IN CHICKENS FED DIETS WITH DIFFERENT PROTEIN LEVEL

ABSTRACT

Two experiments were carried out to evaluate the effect of dietary protein content on body composition, energy retention and energy partition in broiler chickens. Exp. 4.1 was a 2 strain × 2 diet × 2 barrier × 2 sex factorial arrangement and a total of 2,400 Peterson × Arbor Acre and Ross × Hy-Line day-old chicks (males and females) were raised to 42 d of age. Exp. 4.2 was a 2 diet × 4 lighting program factorial arrangement with a total of 2,400 male Peterson × Arbor Acre day-old chicks raised to 42 d in 16 pens. The two diets used in each of the two experiments were the same in composition and similar in nutrient content except for higher protein and amino acid contents in the high-protein (HP) diet than in the low-protein (LP) diet. The results in the two experiments were consistent and confirmed that the HP diet significantly increased body weight gain, feed efficiency, but significantly reduced body lipid content as compared with the LP diet. The birds fed the LP diet consumed higher energy and lower protein amounts than the ones fed the HP diet. A higher proportion of retained energy went to body lipid in the birds fed the LP diet than in those fed the HP diet over the whole feeding period in Exp. 4.2 (0.60 vs. 0.53, respectively). The HP diet significantly decreased energy retained in the chickens as compared with the LP diet. With the increase of body energy retained per kg $W^{0.75}$, an increasing proportion of the total energy retained was directed to body lipids. The results in the current study indicated that the HP diet significantly reduced body lipid accretion, decreased energy retention and directed relatively less of the retained energy to lipid accretion as compared with the LP diet.

Key words: dietary protein, body composition, energy retention, energy partition, broiler

INTRODUCTION

It was reported that a HP diet can significantly affect lipid metabolism in broiler chickens (Kirchgessner et al., 1978; Jackson et al., 1982; Keren-Zvi et al., 1992). Increasing dietary protein content at a constant dietary energy level was reported to reduce hepatic lipogenesis (Yeh and Leveille, 1969; Rosebrough et al., 1988). Kouba et al. (1992) confirmed that a high-protein diet decreased malic enzyme activity of broilers. This effect is independent of the decrease in dietary carbohydrate that accompanied the increased protein level (Leveille et al., 1975; Tanaka et al., 1983).

Kirchgessner et al. (1978) found that broiler chickens deposited less lipid and less energy as the dietary protein/energy ratio increased. The absolute amount of body protein increased with dietary protein level until it reached 24%, and was constant between 24-36% protein (Jackson et al., 1982). Both poor-quality protein (feather meal) and high-quality protein (soybean plus DL-methionine) were equally effective in reducing broiler body lipid deposition (Griffith et al., 1977).

A HP diet can change the partition of absorbed energy, with a greater proportion of energy retained directed to lean tissue (protein) and a decreasing proportion to adipose tissue (Kirchgessner et al., 1978; Jackson et al., 1982). Boekholt et al. (1994) reported that as energy retention increased, fat and protein were retained in a constant energy ratio of 85: 15.

Teeter and Wiernusz (1994) attributed the reduction in energy retention efficiency associated with a HP diet to an increase in the animal's heat production. Contrary findings have also been reported. Several studies reported that high protein diets reduce heat production and increase energy retention efficiency in rats (Tulp et al., 1979; Swick and Gribskov, 1983; Coyer et al., 1987). Since the published results are inconsistent in energy partition between body lipid and protein, and in energy retention as affected by dietary protein level, it is important to do more experiments in this area. The objective of the current study was to further explore the effects of a high-protein diet on chicken body composition, and energy retention and partition.

MATERIALS AND METHODS

Two experiments were conducted to evaluate the effect of dietary protein on body composition and energy retention and partition in broiler chickens. The diets used in the two experiments were similar in nutrient content except for dietary protein and amino acids as listed in Table 4.1. The HP and LP diets contained about 25 and 20% protein (crude protein) in the starter formula for 0-3 wk and about 21 and 16% protein during 3-6 wk, respectively. Laboratory analyses of samples and statistical analysis of data in this study were conducted using the same methods and procedures as used in Chapter III.

Experiment 4.1

Exp. 4.1 is the same experiment as Exp. 3.1 in Chapter III. It was a 2 strain × 2 diet × 2 barrier × 2 sex factorial arrangement. The birds were raised to 42 d of age and body composition of sample birds collected at 3 and 5 wk of age were analyzed in the laboratory. The detailed experimental design and procedures are provided in the Materials and Methods in Chapter III.

Experiment 4.2

Exp. 4.2 incorporated dietary protein levels and lighting programs in a 2 × 4 factorial trial. In this experiment, a total of 2,400 male Peterson × Arbor Acre day-old chicks were randomly assigned to 16 floor pens measuring 24 m^2 in groups of 150 each. Each diet had eight replicate pens with wood shavings litter, a gas brooder, four hanging tube feeders, and two water troughs. The birds were offered the two diets *ad libitum* to 42 d of age.

The four lighting programs were a control (CON), an increasing (INC1), an intermittent (INT) and an increasing-intermittent (INC-INT) lighting program. Lighting intensity was 20 lx for the first 3 d and 5 lx from 4 to 42 d. See details about the lighting programs in Chapter VIII.

Birds were fed *ad libitum* to 42 d of age. Total weight of all chickens in each pen was measured at 3 and 6 wk. Four chickens per pen, i.e., 56 chickens per diet were randomly collected at each of 2, 3, 5 and 6 wk of age. Room temperature control was the same as in Exp. 3.1 (see Materials and Methods in Chapter III). In this experiment, partition coefficients of

stored energy to lipid and protein were calculated as k_f = (carcass lipid, g × 9.2 kcal/g)/(carcass lipids, g × 9.2 kcal/g + carcass protein, g × 5.6 kcal/g).

Table 4.1 Diet composition and nutrient contents (percentage unless stated) in Exp. 4.1 and 4.2

Ingredient	Starter diets		Grower diets	
	Low-protein	High-protein	Low-protein	High-protein
Corn	30	30	35	38
Wheat	31	10	35	12.5
Barley	10	10	12	10
Soybean meal	18	35	12	30
Meat meal	7	8	2	5
Tallow	1.8	4	2	2
DL-Methionine	0.2	0.6	0.1	0.4
L-Lysine. HCl	0.2	0.6	0.1	0.4
Others[1]	1.81	1.81	1.81	1.81
ME (kcal/kg)[2]	2954	2874	3056	2911
Determined analysis				
Exp. 4.1[3]				
Crude protein	19.5	24.5	16.3	21.8
Energy/protein ratio	151	119	187	134
Ether extract	4.1	5.9	3.8	6.2
Crude fiber	3.3	3.8	3.2	3.7
Calcium	0.88	0.98	0.69	0.73
Phosphorus	0.81	0.90	0.66	0.58
Exp. 4.2[3]				
Crude protein	20.4	25.9	16.5	21.0
Energy/protein ratio	145	113	185	137
Ether extract	2.6	5.9	2.9	6.8
Crude fiber	3.5	4.0	3.7	4.2
Calcium	0.95	0.99	0.74	0.83
Phosphorus	0.78	0.86	0.59	0.65

[1] Others included 0.5% vitamin premix, 0.5% mineral premix, 0.5% dicalcium phosphate ($CaHPO_3$), 0.2% limestone, 0.08% Coban (Coccidiostat), and 0.03% Zinc Bacitracin. The vitamin premix supplied per kg of diet with vitamin A 9000 IU; vitamin D_3 1500 IU; vitamin E 10 IU; vitamin K_3 0.5 mg; vitamin B_{12} 0.007 mg, thiamin 0.4 mg; riboflavin 6 mg; folic acid 1 mg, biotin 0.15 mg; niacinamide 35 mg; pyridoxine 4 mg, choline chloride 1,000 mg; and Ethoxyquin 0.125 g. The mineral premix supplied per kg diet with salt 2g; manganese 60 mg, copper 5 mg, zinc 50 mg, selenium 0.1 mg, and iodine 0.35 mg.

[2] Calculated metabolizable energy values.

[3] Exp. 4.1 was a 2 strain × 2 diet × 2 barrier × 2 sex factorial experiment; Exp. 4.2 was a 2 diet × 4 lighting program factorial experiment.

RESULTS

Experiment 4.1

Chickens fed the HP diet grew faster, and converted feed to body weight more efficiently than those fed the LP diet ($p<0.001$, Table 4.2) during each of the age periods. Although their feed intake was lower, the chickens fed the HP diet consumed more protein but less energy ($p<0.001$, Table 4.2) than their counterparts during 0-3, 3-6 and 0-6 wk, due to the higher dietary protein and lower feed intake for the HP diet. There was a significant interaction between diets and sex in body weight at both 3 and 6 wk of age and in protein intake during 0-6 wk. Male chickens fed the HP diet reached higher body weights than those fed the LP diet (2354 vs. 2285 g, respectively), while female chickens fed the HP diet had similar body weights at 6 wk of age as compared with female chickens fed the LP diet (1990 vs. 1987 g, respectively, $p<0.01$). A similar interaction between diet and sex occurred in 3-wk body weight ($p<0.01$). There was a narrower difference in protein intake between male and female birds (20.6 vs. 18.8 g/d, respectively, during 0-6 wk) on the HP diet than on the LP diet (17.0 vs. 15.0 g, respectively, during 0-6 wk). Similar interactions occurred in protein intake during 0-3 and 3-6 wk of age.

Chickens fed the HP diet were significantly lower in carcass lipid, and higher in carcass protein than those fed the LP diet ($p<0.001$) at 3 and 5 wk of age. Birds fed the HP diet contained lower energy than those fed the LP diet ($p<0.001$) during 0-3 and 0-5 wk (Table 4.3).

Experiment 4.2

In this two diet × four lighting program factorial experiment, the average final body weight of the male birds and feed/gain ratio during the whole growth period were 2457 g/bird and 1.805 respectively, which are normal for commercial male birds (Table 4.2). Chickens fed the HP diet obtained significantly higher body weight, and better feed conversion during 0-3, 3-6 and 0-6 wk (Table 4.2). However, chickens fed the HP diet had lower feed intake, lower AME intake but higher protein intake than those fed the LP diet during any of the growth periods (Table 4.2). Basically, the results in Exp. 4.2 were the same as in Exp. 4.1 as to the effect of dietary protein.

Table 4.2 Effect of a high- and a low-protein diet on performance, and feed, energy and protein intake (Exp. 4.1 and 4.2)

		Body Weight, g		Feed intake, g/d/bird			Feed: gain ratio, g/g			AME intake, kcal/d/bird			Protein intake, g/d/bird		
		3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk
Exp. 4.1[1]															
	High-protein	727[a]	2174[a]	45[b]	134[b]	86[b]	1.38[b]	1.98[b]	1.76[b]	121[b]	367[b]	235[b]	11.3[a]	29.3[a]	19.6[a]
	Low-protien	694[b]	2131[b]	46[a]	146[a]	92[a]	1.48[a]	2.12[a]	1.90[a]	137[a]	445[a]	279[a]	9.3[b]	23.8[b]	16.0[b]
	Significance	***	***	**	***	***	***	***	***	***	***	***	***	***	***
	SEM[3]	2	7	0	1	1	0.01	0.02	0.01	1	3	2	0.1	0.2	0.1
	N[4]	612	574	24	24	24	24	24	24	24	24	24	24	24	24
	Diet × sex	**	**										***	***	***
Exp. 4.2[2]															
	High-protein	860[a]	2551[a]	51[b]	150[b]	94[b]	1.34[b]	1.87[b]	1.67[b]	139[b]	410[b]	257[b]	13.3[a]	32.3[a]	21.6[a]
	Low-protein	818[b]	2363[b]	53[a]	167[a]	100[a]	1.46[a]	2.24[a]	1.94[a]	157[a]	509[a]	311[a]	10.8[b]	28.0[b]	18.3[b]
	Significance	**	***	*	**	**	***	***	***	***	***	***	***	**	***
	SEM[3]	7	20	0	4	2	0.03	0.02	0.09	1	10	4	0.1	0.8	0.3
	N[4]	8	8	8	8	8	8	8	8	8	8	8	8	8	8

[ab] Means with different superscripts are significantly different, * $p<0.05$; ** $p<0.01$; *** $p<0.001$.

[1] Exp. 4.1 was a 2 strain × 2 diet × 2 barrier level × 2 sex factorial design. Other significant interactions are reported in Chapter III.

[2] Exp. 4.2 was a 2 diet × 4 lighting program factorial design with male broilers only. The two-way interaction between diet and lighting programs was not significant ($p>0.05$).

[3] SEM: Standard error of mean.

[4] N: number of observations.

Broilers fed the HP diet contained significantly ($p<0.001$) lower body lipid at 2, 3, 5, and 6 wk of age than those fed the LP diet. Even at 2 wk of age, there was great difference in body lipid content between the two diet groups (8.0 vs. 9.2% respectively, for the HP and LP diets). By 6 wk of age, chickens fed the HP diet contained 21.8% less body lipid than those fed the LP diet (12.9 vs. 16.5%, respectively). Generally, body lipid content increased with body weight, but the relationship was curvilinear (Fig. 4.1). At an early age, body lipid content increased dramatically with body weight, while at later stages body lipid content increased slowly with body weight (Fig. 4.1). Chicken body lipid content did not increase much after 5 wk of age. The average body lipid contents at 5 and 6 wk were 14.6 and 14.7% respectively for the male birds in this experiment.

Carcass protein content was significantly higher ($p<0.001$) for birds fed the HP diet. The absolute protein amount deposited in the body for those fed the HP diet was also higher ($p<0.001$) than that for the birds fed the LP diet (Table 4.3). Carcass water was significantly higher ($p<0.001$) for the birds fed the HP diet at 6 wk but not at 3 wk (not listed in Table 4.3).

The absolute and relative amount of calories in the body of birds fed the HP diet was lower at 6 wk as compared with the birds fed the LP diet. The HP diet had less energy partitioned to body lipids than the LP diet (0.53 vs. 0.60 during 0-6 wk, $p<0.001$). Fig. 4.2 depicted the relationship between energy retained as lipid per kg $W^{0.75}$ and total energy retained per kg $W^{0.75}$. As body energy content per kg $W^{0.75}$ increased, increasing energy was retained as lipid. The partition coefficient of retained energy to lipid (k_f) gradually increased with total energy retained per kg $W^{0.75}$ (Fig. 4.3). In other words, as total energy amount increased, a decreasing proportion of the retained energy went to protein. The effect of lighting programs will be reported in Chapter VIII.

DISCUSSION

Protein Intake and Body Weight Gain

Chickens consuming the HP diet had higher body weight at both 3 and 6 wk of age in the two experiments. Jackson et al. (1982) also reported that the chickens fed a diet containing 24%

Table 4.3 Effect of a high- and a low-protein diet on carcass composition and energy partition (Exp. 4.1 and 4.2)

		Carcass lipids					Carcass protein			Carcass energy[5]			k_f[6]	
		%	% 3 wk	% 5 wk	%	g/bird 5 wk	% 3 wk	% 5 wk	g/bird 5 wk	kcal/kg 3 wk	kcal/kg 5 wk	kcal/bird 5 wk		
Exp. 4.1[1]														
	High-protein		10.4[b]	14.7[b]		320[b]	18.4[a]	18.4[a]	406[a]	2008[b]	2404[b]	5220[b]		
	Low- protein		12.4[a]	17.2[a]		368[a]	18.0[b]	17.7[b]	382[b]	2175[a]	2605[a]	5537[a]		
	Significance		***	***		***	***	***	***	***	***	***		
	SEM[3]		0.1	0.2		3	0.1	0.1	2	13	18	37		
	N[4]		120	120		120	120	120	120	120	120	120		
Exp. 4.2[2]		2 wk	3 wk	5 wk	6 wk	6 wk	3 wk	6 wk	6 wk	3 wk	6 wk	6 wk	0-3 wk	0-6 wk
	High- protein	8.0[b]	9.3[b]	13.3[b]	12.9[b]	332[b]	17.6[a]	19.1	491[a]	1886	2285[b]	5875[b]	0.48[b]	0.53[b]
	Low- protein	9.2[a]	10.5[a]	15.9[a]	16.5[a]	393[a]	16.7[b]	18.6	443[b]	1920	2581[a]	6176[a]	0.51[a]	0.60[a]
		*	***	***	***	***	**		***		***	**	***	***
	SEM[3]	0.2	0.3	0.3	0.4	12	0.3	0.3	10	19	36	84	0.01	0.01
	N[4]	32	32	32	32	32	32	32	32	32	32	32	32	32

[ab] Means with a different letter differ significantly, * $p<0.05$; **$p<0.01$; ***$p<0.001$.

[1] Exp. 4.1 was a 2 strain × 2 diet × 2 barrier level × 2 sex factorial design. The interactions of diet and other factors are reported in Chapter III.

[2] Exp. 4.2 was a 2 diet × 4 lighting program factorial design with male broilers only. The two-way interaction between diet and lighting programs was not significant ($p>0.05$).

[3] SEM: Standard error of mean.

[4] N: number of observations.

[5] Carcass energy retained = body fat, g × 9.2 kcal/g + body protein, g × 5.6 kcal/g.

[6] K_f = energy retained as lipids/total energy retained.

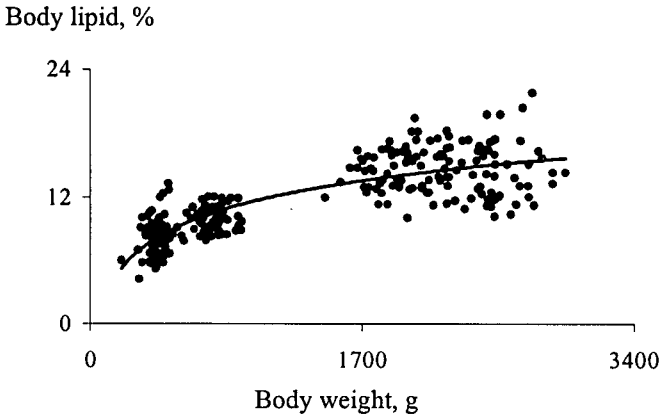

Fig. 4.1 Relationship between body weight and body lipid content (Exp. 4.2)

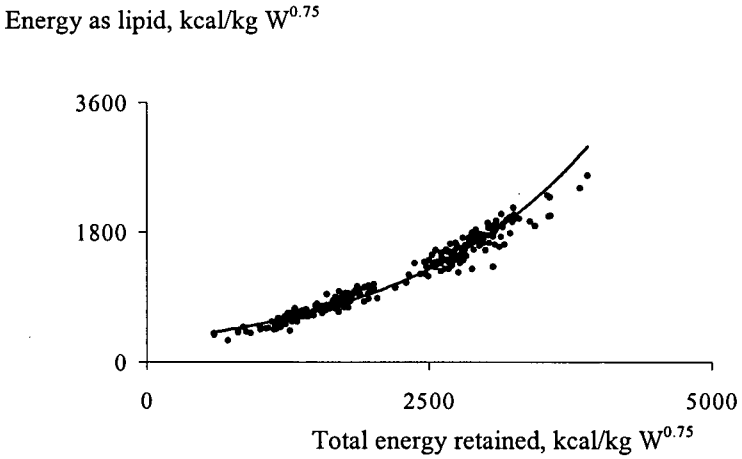

Fig. 4.2 Relationship between energy retained as lipids and total energy retained (Exp. 4.2)

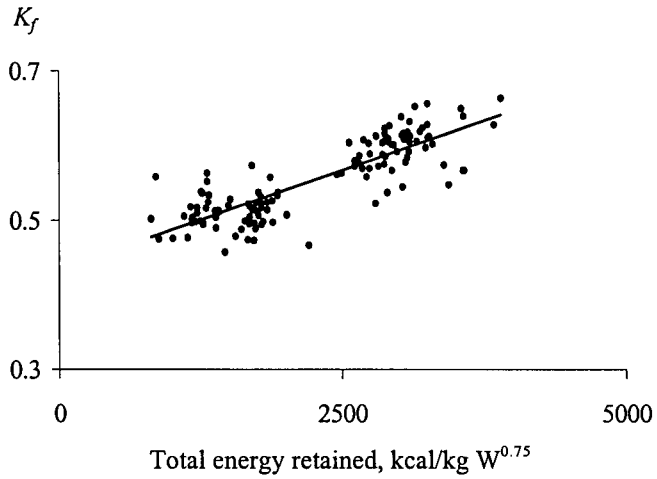

Fig. 4.3 Relationship between energy partition coefficient to lipid (k_f) and total energy retained (Exp. 4.2, for low protein diet only)

protein showed higher body weight at 7 wk of age than those fed a diet containing 20% protein (1766 vs. 1734 g/bird). There are two possible explanations for the body weight difference between the two diets in the current study. One explanation is that the protein in the LP diet was too low (16%) to support the bird's normal growth, resulting in lower body weight. In Exp. 4.2, the difference in final body weight was even more pronounced (2551 vs. 2363 g for the birds fed the HP and LP diets, respectively) between the two diets (Table 4.2). This is probably because the protein deficit was even greater for the male birds fed the LP diet as male birds need higher dietary protein content than females (NRC, 1994). The other explanation is that the high protein intake with the HP diet reduced body lipid and increased lean tissue deposition. Since lean tissue deposition needs less energy than adipose tissue deposition, high protein intake will increase body weight gain. High protein intake itself might also increase muscle protein synthesis (Muramatsu et al., 1987) and stimulate lean tissue (protein) growth (Kirchgessner, 1978) within a given range, resulting in higher body weight compared with low protein intake.

The diet and sex interaction in body weight in Exp. 4.1 indicated that male and female chickens had different responses to dietary protein concentration, i.e., the body weight gain of male (not female) chickens increased when dietary protein content increased. Greater difference in protein intake between male and female birds on the LP diet than on the HP diet may suggest that male birds increase feed intake on the LP diet to meet their higher protein requirement (NRC, 1994) which did not happen on the HP diet.

Body Composition

In both Exp. 4.1 and 4.2, body lipid was significantly lower for birds fed the HP diet (e.g., 14.7%) than for birds fed the LP diet (e.g., 17.2%, Table 4.3). These results are consistent with previous findings (Kirchgessner et al., 1978; Jackson et al., 1982). On a similar dietary energy level basis, a HP diet has been reported to reduce hepatic lipogenesis (Yeh and Leveille, 1969; Rosebrough et al., 1988). The lower hepatic lipogenesis with a HP diet may be responsible for the lower body lipid for birds fed the HP diet in the current experiments.

The results from the two experiments showed that the birds fed the HP diet contained significantly higher body protein compared with the birds fed the LP diet (Table 4.3). The absolute amount of protein deposited in the body for the birds fed the HP diet was also higher

than for the birds fed the LP diet in both experiments (e.g., 406 vs. 382 g/bird at 6 wk of age in Exp. 4.1, Table 4.3). The lower dietary protein content in the LP diet probably failed to meet the bird requirements for maximum potential of protein synthesis. This may explain why the birds fed the LP diet showed relatively and absolutely lower body protein content. Jackson et al. (1982) also reported that the birds fed a 16% protein diet deposited less body protein than those fed diets containing 20-36% protein. The protein retention efficiency decreased with dietary protein level in both of the current experiments. Body lipid content increased dramatically with body weight during 2-3 wk, and slowly thereafter (Fig. 4.1).

Energy Retention and Feed Efficiency

Chickens fed the HP diet also had lower feed/gain ratios (Table 4.2). The change of feed conversion efficiency was due mainly to body composition change. Birds fed the HP diet had lower relative and absolute carcass energy content than birds fed the LP diet, due to the lower fat content in the birds fed the HP diet. The lower the energy content per unit weight, the lower the feed/gain ratio as discussed before.

It was reported that body lipid synthesis utilizes feed energy more efficiently than body protein synthesis (MacLeod and Geraert, 1988; Larbier and Leclercq, 1994). In other words, more protein deposition and less lipid deposition will result in lower energy retained in the body. In both Exp. 4.1 and 4.2, the birds fed the HP diet deposited greater amounts of protein and lower amounts of lipids, and therefore lower energy in the body than birds fed the HP diet, e.g., 6176 vs. 5875 kcal/bird at 6 wk, respectively, in Exp. 4.2. Birds fed the high-protein diet might have higher rates of heat production (MacLeod, 1991).

Energy Retained in Lipids and Total Energy Retention

Boekholt et al. (1994) reported that as energy retention increased, fat and protein were retained in a constant energy ratio of 85: 15. They observed a linear relationship between energy retained as lipids and total energy retained expressed as kJ/kg metabolic body weight ($W^{0.75}$). However, in the current study, the relationship between energy retained as body lipids and total energy retained is an exponential curve (Fig. 4.1). As the body energy content per kg $W^{0.75}$

increased, increasing energy was stored as lipids. Fig. 4.3 more clearly shows the relationship between the proportion of energy partition to lipids and total energy retained. As total energy retained expressed per kg $W^{0.75}$ increased, an increasing proportion of retained energy was directed into body lipids. Jackson et al. (1982) also reported that as total energy retention increased, the energy retained in the protein did not change, but the energy retained in the fat increased gradually. In other words, the proportion of retained energy in protein out of total retained energy decreased gradually rather than remaining constant with the total energy retained. Our results strongly support those of Jackson et al. (1982).

Disadvantages of a High Protein Diet

The HP diet increased chicken body weight gain, reduced chicken body lipid content, and also improved feed efficiency. However, the HP diet can increase nitrogen in the excreta and possibly lead to environmental pollution (Deschepper and De Groote, 1995). In addition, the HP diet is also more costly and could be considered to be a waste of precious protein sources. Therefore, a HP diet is not recommended for commercial use in the broiler industry. Instead, an LP diet with improved amino acid balance by supplementing synthetic amino acids is a sound approach to minimizing nitrogen excretion (Leeson and Summers, 1997; Ibrahim, 1997).

CONCLUSION

A HP diet significantly reduced body lipid content as well as increased body weight gain and improved feed efficiency as compared with an LP diet. It was found that the HP diet significantly decreased energy retention in the body. As body energy retention increased, more and more retained energy was directed to body lipids.

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CHAPTER V

EFFECT OF AMINO ACID PROFILES WITH A LOWER DIETARY PROTEIN LEVEL ON THE PERFORMANCE AND BODY COMPOSITION OF BROILER CHICKENS

ABSTRACT

Poultry producers may in the future be required to use low-protein diets supplemented with amino acids to reduce nitrogen release to the environment. However, the literature suggests that these diets may increase the lipid content of broilers. The objectives of this study were to study the possibility of using a low-protein diet with an alternate amino acid profile coupled with 4-phase feeding to maintain performance and body composition. Four diets were compared using 1,440 male broiler chicks in a single-factor design. The control diet was based on NRC (1994) recommendations with three phases (0 to 3, 3 to 6 and 6 to 7 wk of age). The amino acid composition in the three test diets (T1, T2, and T3) was based on maintenance amino acid requirements, carcass and feather amino acid profiles, and amino acid digestibility of feed ingredients. The three test diets were formulated on a digestible amino acid basis and had four phases, i.e., 0 to 2, 2 to 4, 4 to 6, and 6 to 7 wk of age. The three test diets contained similar protein level which was 1.2 -2.2 percentage points lower than the control depending on the phase. The lysine, methionine, methionine + cystine, and threonine in T3 and T2 were 11-25% and 6-15% higher than in T1 respectively, depending on amino acid and phase. Birds fed the four diets obtained similar body weight gain, feed conversion, body lipid and protein content at each phase and breast yield at 6 wk. Furthermore, the chickens fed the test diets reduced nitrogen excretion by 7 to 16% compared with those fed the control diet. In terms of nitrogen excretion/retention, Diet T1 was better than the other three diets. This experiment indicated that the protein and amino acid balance in Diet T1 with 4-phase feeding is able to maintain growth performance, body composition, and breast meat yield and to reduce nitrogen excretion by 7-16% as compared with the control diet.

Key words: protein, amino acid balance, nitrogen excretion, body composition, broiler

INTRODUCTION

In broilers only 40 to 60% of feed nitrogen is used for maintenance and production (Blair et al., 1999 and Chong, 1999). The remainder is excreted. Environmental nitrogen pollution originating from animal production has become a major problem (Deschepper and De Groote, 1995), particularly in regions of intensive animal production. Application of excess manure to the soil causes leaching, and subsequent run-off may cause eutrophication (Schutte, 1994).

To improve nitrogen retention and reduce nitrogen excretion, two major measures have been suggested and tested: (1) reduction of dietary protein content and improvement of amino acid balance and; (2) multi-phase feeding. Some researchers reported that maximum performance cannot be achieved by feeding broilers low protein diets supplemented with synthetic amino acids (Uzu, 1982; Jensen, 1991). Other researchers have reported that it is possible to achieve optimal performance using a low-protein diet supplemented with synthetic amino acids (Summers and Leeson, 1985; Schutte, 1987; Parr and Summers, 1991). The discrepancy is probably because the protein content in the low-protein diets was different, and some low-protein diets may need to be supplemented with other limiting amino acids besides lysine and methionine, and/or with non-essential amino acids (Schutte, 1994). A concurrent problem was that chicken carcass fat increased with the reduction of dietary protein level (Parr and Summers, 1991; Moran et al., 1992).

Improvement of dietary amino acid balance not only enhances body protein accretion but also depresses body fat synthesis. Yeh and Leveille (1969) and Rosebrough et al. (1988) reported that an improvement in dietary protein quality (e.g., addition of the limiting amino acid lysine to a low protein diet) depressed *de novo* lipogenesis in chicks. The reduction in body fat and increase in body protein that occurs in response to supplemental limiting amino acids become obvious once dietary protein level is reduced (Mendonca and Jensen, 1989). Grisoni et al. (1991) found that when dietary lysine was increased gradually from 0.7 to 1.1%, abdominal fat was reduced gradually from 6.4 to 4.0%. Pack and Schutte (1992) found that the amount of methionine needed to optimize feed conversion and body composition is greater than that needed to maximize body weight. Blair et al. (1999) formulated three diets containing 18% protein and 3200 kcal metabolizable energy/kg, but different levels of dietary essential amino acids (lysine, methionine + cystine, threonine, and tryptophan) changing from 90, to 100 or 110% of the

control level. They observed that the body fat content of broiler chickens fed the three diets gradually decreased from 16.8, to 16.6 and 16.1% respectively at 6 wk of age. They even observed a more pronounced reduction of the body fat content with an increase of the four essential amino acids at 3 wk of age in another experiment with broilers (Blair et al., 1999). Therefore, it was hypothesized that body fat in chickens fed a low-protein diet might not be increased when dietary amino acid balance was improved and limiting amino acids were supplemented.

Multi-phase feeding is another measure to reduce nitrogen excretion. The requirement of the chicken for dietary protein content gradually decreases with age (NRC, 1994). Currently, NRC (1994) recommended three feeding phases for broiler chickens, i.e., a starter phase (0-3 wk), a grower phase (3-6 wk), and a finisher phase (6-8 wk). In practice, dietary protein content is adjusted down suddenly, e.g., from 23% in the starter phase to 20% in the grower phase. Therefore, the dietary protein content may be excessive at late previous phase and too low at early subsequent phase. Theoretically, the greater number of feeding phases, the better the dietary protein utilization, but too many feeding phases are not practical in broiler production. A four-phase feeding program suggested for broiler chickens (Schutte, 1994) was assumed to minimize the excess of dietary protein without affecting performance.

The objective of the current study was to study the effects of low protein diets with different amino acid profiles coupled with four feeding phases on the performance and carcass composition of broiler chickens.

MATERIALS AND METHODS

Experiment 5.1

Diets and experimental design

This was a co-operative study with Heartland Lysine Inc, Illinois, USA. A single-factor complete randomized design was used to arrange four diet treatments. The main ingredients were local wheat and canola meal, plus soybean meal. A wheat enzyme (Avizyme-TX, mainly microbial xylanase) supplied by Finfeeds International, UK was added to each diet at 0.1% to hydrolyze xylans in wheat. The ingredients were analyzed for protein and amino acid content

before formulating the diets. The control diet was formulated on a total amino acid basis (NRC, 1994). The control diet had three phases, 0 to 3, 3 to 6 and 6 to 7 wk.

The three test diets (Diet T1, T2 and T3) contained the same protein level, but were formulated to be 1.2 -2.2 percentage points lower than the control diets, depending on the growing phase. The preset minimum digestible essential amino acid requirements and the ratios of digestible essential amino acids to digestible lysine (Table 5.1) were developed for the three test diets from maintenance amino acid requirements (Leveille and Fisher, 1959 and 1960; Leveille, et al., 1960), and carcass and feather amino acid profiles (Heartland Lysine, Inc., Illinois). The essential amino acid contents in Diets T2 and T3 were formulated to be 8-12% and 16-23% higher than in Diet T1 respectively depending on the growing phase.

The three test diets (T1, T2 and T3) were formulated on digestible amino acid basis, using the amino acid digestibility data from Heartland Lysine, Inc. and the amino acid contents in the ingredients. The growing period for the three test diets was divided into four phases, i.e., pre-starter, 0-2 wk; starter, 2-4 wk; grower, 4-6 wk, and finisher, 6-7 wk. The diet composition is listed in Table 5.2. Synthetic L-lysine·HCl (98.5%), DL-methionine (99%) and L-threonine (98%) were used to supplement dietary lysine, methionine and threonine, respectively. The true digestibility of the synthetic amino acids was assumed to be 100% (Chung and Baker, 1992). All the diets were supplemented with 0.6% celite (Celite^{TM}, a diatomite product, Food Chemicals Codex Grade, Celite Corp., Lompar, CA 93436) as an acid-insoluble ash marker used to determine dietary AME and nitrogen retention. All the diets were analyzed for protein and amino acid content.

Broiler experiment

1,440 day-old male Peterson × Arbor Acre chicks were randomly assigned to 24 floor pens, with 60 birds per pen. The four diets were randomly assigned to the 24 pens, six replicate pens per diet. The chickens were allowed to feed and drink freely. Feed consumption was measured for each pen for 0 to 2, 2 to 4, 4 to 6 and 6 to 7 wk. About half of the chickens in a pen were weighed individually at each of 2, 4, 6, and 7 wk of age. Two chickens per pen and 12 chickens for each diet at each of 2, 4, 6, and 7 wk of age were randomly collected, weighed and scanned by TOBEC (Appendix I) by the same procedure as in Exp. 3.1. After scanning, the birds were killed, the head, neck, wings and legs removed as in Exp. 3.1, and were weighed,

Table 5.1 Pre-set minimum digestible essential amino acid content[1] (%) of diets and their ratios to lysine (%) in Exp. 5.1

	Diet T1				Diet T2				Diet T3				Control diet		
	0-2 wk	2-4 wk	4-6 wk	6-7 wk	0-2 wk	2-4 wk	4-6 wk	6-7 wk	0-2 wk	2-4 wk	4-6 wk	6-7 wk	0-3 wk	3-6 wk	6-7 wk
Minimum digestible essential amino acid contents															
Lysine	1.06	0.97	0.88	0.75	1.14	1.06	0.97	0.84	1.23	1.14	1.06	0.92	0.98	0.87	0.75
Methionine	0.43	0.39	0.35	0.28	0.49	0.46	0.42	0.35	0.56	0.52	0.50	0.41	0.41	0.28	0.24
Met + Cys	0.76	0.71	0.66	0.57	0.82	0.78	0.73	0.64	0.88	0.84	0.80	0.70	0.69	0.52	0.46
Threonine	0.72	0.66	0.62	0.54	0.76	0.72	0.68	0.60	0.82	0.78	0.74	0.66	0.67	0.63	0.41
Tryptophan	0.24	0.22	0.21	0.19	0.24	0.23	0.21	0.19	0.24	0.22	0.21	0.19	0.23	0.19	0.15
Arginine	1.16	1.06	0.94	0.79	1.25	1.16	1.04	0.89	1.35	1.24	1.13	0.98	1.14	1.00	0.91
Histidine	0.34	0.31	0.29	0.25	0.36	0.34	0.32	0.28	0.39	0.37	0.35	0.31	0.32	0.29	0.24
Isoleucine	0.68	0.61	0.55	0.46	0.74	0.67	0.6	0.51	0.79	0.72	0.66	0.56	0.72	0.66	0.56
Leucine	1.16	1.07	0.99	0.84	1.25	1.17	1.09	0.94	1.35	1.25	1.19	1.03	1.09	0.99	0.85
Phenylalanine	0.63	0.58	0.51	0.42	0.68	0.64	0.56	0.48	0.74	0.68	0.61	0.52	0.66	0.59	0.51
Valine	0.76	0.70	0.62	0.52	0.82	0.76	0.69	0.59	0.88	0.82	0.75	0.64	0.79	0.72	0.62
Essential amino acid/lysine ratios															
Lysine	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
Methionine	40	40	39	39	39	40	39	38	40	40	39	38	47	39	39
Met + Cys	72	73	75	76	72	74	75	76	72	74	75	76	84	74	72
Threonine	67	68	70	72	67	68	70	73	67	68	70	72	71	72	78
Tryptophan	17	16	17	17	17	17	16	17	17	17	17	17	18	18	19
Arginine	109	109	107	105	110	109	107	106	110	109	107	107	118	114	122
Histidine	32	32	33	33	32	32	33	33	32	32	33	34	33	33	33
Isoleucine	64	63	63	61	65	63	62	61	64	63	62	61	75	75	75
Leucine	109	110	113	112	110	110	112	112	110	110	112	112	113	113	114
Phenylalanine	59	60	58	56	60	60	58	57	60	60	58	57	68	68	68
Valine	72	72	70	69	72	72	71	70	72	72	71	70	82	82	83

[1] Minimum digestible essential amino acid contents in the test diets were based on maintenance amino acid requirements (Leveille and Fisher, 1959 and 1960) and carcass and feather amino acid profiles (Heartland Lysine, Inc., Illinois). The essential amino acid contents in Diet T2 and T3 were set to be 8-12% and 16-23% higher than in Diet T1, respectively, depending on the growing phase. The contents of digestible essential amino acids in Diet T1 were set to be higher than those in the control diet (NRC, 1994). The digestible essential amino acid contents in the control diet were based on the essential amino acid digestibility data from Heartland Lysine and the essential amino acid requirements in NRC (1994).

Table 5.2 Diet composition and nutrient analyses (percentage unless stated) in Exp. 5.1

Diet	Diet T1				Diet T2				Diet T3				Control diet		
	0-2 wk	2-4 wk[2]	4-6 wk	6-7 wk	0-2 wk	2-4 wk[2]	4-6 wk	6-7 wk	0-2 wk	2-4 wk[2]	4-6 wk	6-7 wk	0 -3 wk	3 -6 wk	6 -7 wk
Wheat	55.0	59.8	64.6	70.7	43.0	58.4	63.8	70.4	37.4	57.6	65.2	70.6	51.8	60.2	66.7
Corn	--	--	--	--	10.4	--	--	--	14.7	1.9	--	--	--	--	--
Tallow	8.0	7.6	7.3	6.8	8.0	7.8	7.4	6.8	8.0	7.4	6.9	6.8	8.0	7.4	7.2
Soybean meal	26.5	19.7	15.3	11.2	25.2	20.7	15.8	11.1	26.3	22.0	19.0	10.7	34.1	19.8	15.0
Canola meal	5.3	8.0	8.0	7.0	8.0	8.0	8.0	7.0	8.0	5.6	3.6	7.0	0.9	8.0	7.0
Dicalcium phosphate	1.63	1.36	1.11	0.87	1.63	1.36	1.11	0.87	1.64	1.38	1.14	0.87	1.63	1.09	0.85
Limestone	1.35	1.37	1.40	1.31	1.32	1.36	1.40	1.31	1.31	1.38	1.44	1.31	1.38	1.39	1.30
DL-Methionine	0.14	0.12	0.10	0.04	0.21	0.18	0.16	0.11	0.27	0.25	0.25	0.17	0.17	0.07	0.03
L-Lysine·HCl	0.07	0.12	0.13	0.10	0.18	0.20	0.23	0.22	0.27	0.30	0.32	0.34	--	--	--
L-Threonine	--	--	0.01	--	0.04	0.05	0.07	0.07	0.09	0.11	0.13	0.13	--	--	--
Others[1]	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03	2.03
Determined analyses (on air-dry basis)															
AME, kcal/kg	2600	3150	3169	3179	2696	3150	3138	3130	2590	3150	3095	3169	2775	3225	3078
Crude protein	21.1	19.9	18.4	16.8	21.2	20.4	18.5	16.7	21.2	20.4	18.7	16.5	23.9	19.5	17.9
Ether extract	10.5	9.6	9.1	8.0	9.7	9.7	8.5	7.4	10.8	9.3	8.1	7.1	9.0	8.7	6.8
Calcium	1.2	1.0	0.9	0.7	1.0	0.9	0.9	0.7	1.2	1.0	0.8	0.7	0.9	0.8	0.8
Phosphorus	0.73	0.69	0.60	0.52	0.70	0.69	0.64	0.56	0.82	0.68	0.67	0.50	0.68	0.60	0.57
Lysine	1.19	1.11	0.98	0.89	1.27	1.21	1.10	0.94	1.39	1.28	1.16	1.02	1.12	1.05	0.86
Methionine	0.59	0.56	0.52	0.44	0.67	0.63	0.60	0.55	0.80	0.71	0.65	0.55	0.56	0.39	0.33
Met + Cys	0.99	0.94	0.89	0.80	1.08	1.01	0.96	0.91	1.14	1.08	1.01	0.90	0.94	0.71	0.63
Threonine	0.85	0.78	0.72	0.65	0.92	0.84	0.79	0.69	0.94	0.90	0.83	0.77	0.81	0.76	0.65
Tryptophan	0.31	0.26	0.28	0.26	0.29	0.26	0.25	0.25	0.30	0.26	0.25	0.22	0.27	0.23	0.18
Calculated digestible essential amino acid contents (on air-dry basis)															
Lysine	1.06	0.97	0.88	0.75	1.14	1.06	0.97	0.84	1.23	1.14	1.06	0.92	0.98	0.87	0.75
Methionine	0.43	0.39	0.35	0.28	0.49	0.46	0.42	0.35	0.56	0.52	0.50	0.41	0.41	0.28	0.24
Met + Cys	0.76	0.71	0.66	0.57	0.82	0.78	0.73	0.64	0.88	0.84	0.80	0.70	0.69	0.52	0.46
Threonine	0.72	0.66	0.62	0.54	0.76	0.72	0.68	0.60	0.82	0.78	0.74	0.66	0.67	0.63	0.41
Tryptophan	0.24	0.22	0.21	0.19	0.24	0.23	0.21	0.19	0.24	0.22	0.21	0.19	0.23	0.19	0.15

[1] Include 0.60% Celite, 0.50% vitamin premix, 0.50% mineral premix, 0.25% iodized salt, 0.1% Avizyme-TX, and 0.075% Coban-60. The vitamin and the mineral premixes were the same as in Table 3.1.

[2] Nutrient contents for Diets T1, T2 and T3 during 2-4 wk were calculated as the feed samples were missing.

bagged and frozen for later analyses. At 6 wk of age, the breast meat of each sampled carcass was deboned and weighed. The breast meat weight was expressed as a relative weight of the body weight (%) and absolute weight (g). After weighing, the breast meat was put back to the carcass before being bagged and frozen.

Balance study

At the same time as the feeding trial, another 144 day-old male Peterson × Arbor Acre chicks were used in a balance study to measure dietary apparent metabolizable energy and nitrogen retention efficiency. The 144 chicks were randomly assigned to 24 brooder cages, six chicks in each cage measuring about 100 × 70 × 40 cm. Each diet had six replicate cages. The marker method (Scott and Boldaji, 1997) was used with Celite as insoluble ash marker. At each of 2, 4, 6, and 7 wk of age, feed and excreta samples were collected for two successive days. Excreta samples of each cage were pooled and dried at 65 C in a drying oven and ground through a 40 # sieve. Dry matter of feed and excreta samples were determined at 105 C. The feed samples and the 65 C-dried excreta were further analyzed in the lab.

Laboratory analyses and statistical analyses

The frozen chicken carcass samples were prepared and analyzed as in Exp. 3.1 in Chapter III. Gross energy, nitrogen, crude fat, calcium and phosphorus contents were analyzed as in Chapter III. Dietary amino acids were analyzed by the laboratory of Heartland Lysine Inc. using the methods outlined by Spackman et al. (1958). Hydrolysates for the determination of methionine as methionine sulfone and cysteine/cystine as cysteic acid were prepared by performic oxidation of the protein followed by the 6N HCl hydrolysis (Moore, 1963), whereas feed indredients were subjected to alkaline hydrolysis and high performance liquid chromatography for tryptophan determination (Jones et al., 1981). Acid-insoluble ash in both diet and excreta samples was analyzed using the method of Vogtmann et al. (1975). The AME values were determined using the following equation (Scott and Boldaji, 1997):

$$\text{AME (kcal/kg diet on dry matter basis)} = GE_{diet} - GE_{excreta} \times (IA_{diet}/IA_{excreta})$$

Where GE = gross energy, kcal/kg on dry matter basis; IA= content (%) of acid insoluble ash, on dry matter basis. Nitrogen retention was calculated using the following equation:

$$\text{Nitrogen retention (\%)} = 100 - 100 \times (IA_{diet} \div IA_{excreta}) \times (N_{excreta} \div N_{diet})$$

Where N = nitrogen content (%) on dry matter basis.

Effect of the diets on body weight, feed intake, feed conversion, nitrogen retention, excreta nitrogen content, body fat, breast meat yield and percentage were analyzed using analysis of variance for a complete randomized design. Any significant differences between the four diets were further compared using Duncan's multiple range test (SAS Institute Inc., 1996).

RESULTS

Dietary AME, Protein and Amino Acid Contents

Dietary AME and protein contents were very close to the calculated except the three test pre-starter diets for 0-2 wk and the control starter diet for 0-3 wk which had much lower dietary AME levels than calculated (2590-2775 vs. 3150 kcal/kg, respectively, Table 5.2). Dietary protein contents in the three test diets were similar but were 1.2-2.2 percentage points lower than the control diet. Except the first three limiting amino acids (lysine, methionine, and threonine) that just met the preset requirements, all other dietary essential amino acids exceeded the preset requirements listed in Table 5.1. Therefore, only lysine, methionine, methionine + cystine, threonine contents were tabulated in Table 5.2. In this report, just the actually analyzed lysine, methionine, methionine + cystine, threonine contents were used in comparison and discussion. Lysine, methionine, methionine + cystine, and threonine in Diet T2 and T3 were 6-15% and 11-25% higher than in Diet T1, respectively (Table 5.2). The three test diets contained lysine, methionine, methionine + cystine, and threonine than the control diet (Table 5.2).

Dietary Digestibility, Nitrogen Retention and Nitrogen Excretion

The digestibility of dietary dry matter was similar among the four diets during 0-2, 4-6, or 6-7 wk of age. The T1, T2 and T3 dietary samples for 2-4 wk of age were unfortunately misplaced and not available for analysis. The dry matter digestibility gradually increased with age (from 60 to 71%). However, the nitrogen retention efficiency during 0-2 wk was different among the diets (Table 5.3). Diets T1, T2 and T3 had gradually reducing nitrogen retention efficiency in that order, each significantly different from the other during 0-2 wk ($p<0.05$). The

Table 5.3 Effect of dietary protein content and amino acid balance on performance and nitrogen excretion

Age (wk)	Diet	Body weight[1], g	Weight gain, g	Feed: gain ratio	Feed intake, g	N retention[2] %	DM digestibility[2] %	Excreta N, % DM[2]
0-2	T1	415	370	1.27	470	57.2[a]	59.7	4.0[c]
	T2	419	374	1.25	468	48.2[c]	60.2	4.9[b]
	T3	424	379	1.25	473	38.8[d]	58.3	5.1[a]
	Control	416	371	1.26	469	52.8[b]	62.0	5.4[a]
	SEM[3]	6	6	0.01	7	1.2	1.1	0.1
	N[3]	190	6	6	6	6	6	6
2-4	T1	1268	853	1.65	1411	n.a.	n.a.	3.8[c]
	T2	1267	848	1.65	1396	n.a.	n.a.	4.2[b]
	T3	1265	842	1.64	1384	n.a.	n.a.	4.1[bc]
	Control[4]	1284	868	1.64	1422	n.a.	n.a.	4.6[a]
	SEM	8	9	0.01	14			0.1
	N	186	6	6	6			6
4-6	T1	2400	1134	1.92	2173	48.2	67.7	5.0
	T2	2364	1097	1.93	2114	45.7	68.1	5.5
	T3	2345	1080	1.96	2116	49.2	68.7	5.4
	Control	2413	1129	1.92	2171	50.0	69.0	5.8
	SEM	18	29	0.03	31	3.8	1.0	0.3
	N	187	6	6	6	6	6	6
6-7	T1	2885	485	2.44	1185	51.4	70.0	4.8
	T2	2859	495	2.33	1151	55.9	71.1	4.5
	T3	2837	492	2.26	1110	52.9	71.3	4.8
	Control	2830	417	2.58	1075	54.2	70.9	5.0
	SEM	24	37	0.11	36	1.9	0.8	0.2
	N	175	6	6	6	6	6	6
0-7	T1	2885	2840	1.81	5106	52.3[a]	65.8	4.4[c]
	T2	2859	2814	1.81	5052	49.9[ab]	66.5	4.8[b]
	T3	2837	2792	1.80	4988	47.0[b]	66.1	4.9[ab]
	Control	2830	2785	1.82	5049	52.3[a]	67.3	5.2[a]
	SEM	24	31	0.01	62	1.7	0.9	0.1
	N	175	6	6	6	6	6	24

[abcd] Means in each period and a column with different superscripts differ significantly ($p<0.05$).

n.a: not available as the dietary samples were missing.

[1] Body weight was measured at 2, 4, 6, and 7 wk of age.

[2] N: nitrogen; DM: drymatter.

[3] SEM: standard error of mean; N: number of observations.

[4] Control starter and control grower were used in 3 and 4 wk, respectively.

nitrogen retention efficiency of the control diet was significantly ($p<0.05$) lower than that of Diet T1, but significantly ($p<0.05$) higher than Diets T2 and T3 during 0-2 wk. During 4-6 and 6-7 wk, all the diet groups showed similar nitrogen retention efficiency. At all the ages, excreta nitrogen content of the control birds was significantly ($p<0.05$) or non-significantly ($p>0.05$) higher than that of other bird groups (Table 5.3). On average, the excreta nitrogen content for birds fed Diets T1, T2 and T3 was 16, 9, and 7% lower respectively than that of the control ($p<0.05$, Table 5.3).

Growth Performance and Body Composition

The broilers reached 2381 g and 2853 g at 6 and 7 wk of age respectively. There were no significant differences ($p>0.05$) in body weight, weight gain, feed intake, feed conversion (Table 5.3), relative percentage and absolute amount of carcass fat and protein in any period or 6-wk breast yield (Table 5.4) among the four diet treatments.

DISCUSSION

Growth Performance

The determined AME contents for pre-starter Diets T1, T2 and T3 (0-2 wk) and the control starter diet (0-3 wk) were much lower than calculated (3150 kcal/kg). This is probably because the activity of digestion enzymes in the intestinal tract of young chicks is still low at early ages and arabinoxylans in wheat significantly inhibit the digestibility of nutrients at early ages as will be discussed in Chapter VI. Although microbial arabinoxylanase was added, the enzyme did not really bring the AME level to that calculated.

AME intake was similar among the four diets for all the phases. Protein intake for the control birds was higher than for the birds fed the test diets due to the higher protein content in the control diet. The amino acid intake was proportional to the dietary amino acid content, since the feed intake was similar. The chickens on the low-protein diets (T1, T2 and T3) had similar body weight, feed intake, and feed conversion at any age as compared with the control birds.

Table 5.4 Effect of dietary protein and amino acid balance on body composition and breast yield

Age (wk)	Diet	Carcass water[1], %	Carcass fat[1], %	Carcass protein[1], %	Fat retained[1], g	Protein retained[1], g	Breast yield[2], g	Breast / weight[2], %
0-2	T1	70.4	8.8	18.7	32	70		
	T2	70.3	9.4	18.3	35	69		
	T3	70.2	9.3	18.3	35	70		
	Control	70.5	8.8	18.6	32	70		
	SEM[3]	0.3	0.4	0.1	2	1		
	N[3]	12	12	12	6	6		
2-4	T1	65.5	13.0	19.2	128	166		
	T2	65.6	13.1	19.1	127	165		
	T3	65.0	13.7	19.1	134	164		
	Control[4]	65.6	12.9	19.3	129	170		
	SEM	0.4	0.5	0.2	6	3		
	N	12	12	12	6	6		
4-6	T1	62.1	15.5	20.1	207	239	337	14.3
	T2	62.5	15.2	19.8	193	226	323	14.0
	T3	62.2	15.7	19.7	195	220	335	14.2
	Control	62.2	15.0	20.3	196	242	335	14.2
	SEM	0.6	0.7	0.2	24	14	11	0.1
	N	12	12	12	6	6	24	24
6-7	T1	61.5	16.4	19.6	107	83		
	T2	62.1	15.8	19.7	92	95		
	T3	62.2	15.7	19.7	74	97		
	Control	61.7	16.2	19.7	97	88		
	SEM	0.4	0.6	0.3	23	16		
	N	12	12	12	6	6		
0-7	T1	61.5	16.4	19.6	475	558		
	T2	62.1	15.8	19.7	448	555		
	T3	62.2	15.7	19.7	438	551		
	Control	61.7	16.2	19.7	456	560		
	SEM	0.4	0.6	0.3	23	21		
	N	12	12	12	6	6		

[1] Body composition was measured at 2, 4, 6, and 7 wk of age.
[2] Breast meat yield was measured at 6 wk of age as g/bird and per cent of live body weight.
[3].SEM: standard error of mean; N: number of observations.
[4] Control starter and control grower were used in 3 and 4 wk, respectively.

This indicated that the lower dietary protein level with an alternate essential amino acid profile, plus the four-phase feeding for the test diets was successful and practical without affecting growth performance of broilers. These results in the current study clearly showed: 1) the low-protein level used in the test diets is adequate in terms of growth performance compared with the high protein content in the control diet; 2) the higher lysine, methionine, methionine + cystine and threonine in Diets T2 and T3 compared with Diet T1 are not necessary. The results indicate that Diet T1 is better than the other three diets since excreta nitrogen content and dietary essential amino acid contents of Diet T1 were lower than those of Diets T2 and T3.

Body Composition

The most important result in this study was that the body fat content of birds fed the low protein diets (Diets T1, T2 and T3) was not increased ($p>0.05$) as compared with that of the control birds. This result indicated that the low protein diets with an alternate amino acid balance were successful in maintaining bird body fat content. Dietary protein level can markedly alter body lipid deposition. Based on the published results (Kirchgessner et al., 1978; and Jackson et al., 1982), every percentage point decrease in dietary protein content within the range of 16 to 28% can result in an increase of 0.4 percentage points in body fat content of broiler chickens. In the current trial, the dietary protein levels in the test diets were 1.2 to 2.2 percentage points lower than the corresponding control diet. Therefore, the birds fed the test diets were expected to be about 1 percentage point higher in carcass fat content than the control birds. However, the body fat content of the birds fed the test diets was similar to that of the control birds ($p>0.05$) at all the ages. The results indicate that the low protein diets with different amino acid profiles prevented the increase of body lipids resulting from the low dietary protein content. The hypothesis made in the Introduction of this chapter proved to be correct.

Deschepper and De Groote (1995) observed that a low-protein diet (21%/20% protein for 0-3/3-6 wk) with similar essential and non-essential amino acids, significantly increased 6-wk body fat content (14.4 vs. 12.3%, $p<0.05$) as compared with the control diet (23/21% protein for the corresponding periods). This is in contrast to the current results. The reason for this difference is probably that the test diets in the current study were different in amino acid balance, while the low-protein diet in Deschepper and De Groote (1995) was similar in amino acid

balance and contents to the control diet. The different amino acid balance and contents in the current test diets might reverse the lipids-increasing effect of the low protein level.

The body protein content and breast yield were similar among the four bird groups during each of four phases in the present experiment. This showed that the lower protein plus higher essential amino acids in Diet T1 did not reduce body protein synthesis compared with the control. The even higher essential amino acids in Diets T2 and T3 did not increase absolute and relative body protein content and breast yield. Animal's requirement for protein is fundamentally the requirement for amino acids. In the current study, dietary essential amino acids in the low-protein diets (T1, T2 and T3) met or exceeded the requirements of broilers for amino acids. Excessive dietary protein and amino acids do not increase body protein deposition, especially when the excessive dietary amino acids are not well balanced relative to bird requirements (Schutte, 1994). This may explain why the higher dietary protein level in the control diet and higher lysine, methionine, methionine + cystine, and threonine level in the Diets T2 and T3 did not increase body protein content as compared with Diet T1.

Nitrogen Excretion and Retention

During each feeding phase and overall, excreta nitrogen content from the control birds was higher ($p<0.05$, in some phases) than from the birds fed the test diets due probably to the higher protein concentration in the control diet. Similar results were obtained in a number of studies (Blair et al., 1976; Parr and Summer, 1991; Ibrahim, 1997; Blair et al., 1999).

The average excreta nitrogen excreted by the birds fed the three test diets was in the order of Diet T1< T2 < T3 over 0-7 wk of age. This left the nitrogen retention efficiency in a decreasing order for Diets T1, T2 and T3 since the three diets contained similar protein. This result further demonstrated that the higher essential amino acid contents in Diets T2 and T3 were excessive, thus resulting in the decreased nitrogen retention in birds fed these two diets. The non-significant difference in nitrogen retention during 4-6 and 6-7 wk of age is probably because the older birds had less sensitivity to the difference in dietary amino acid balance than the younger birds. Ibrahim (1997) reported that at 21% dietary protein level, essential amino acid at 110, 100 and 90% of the control level resulted in excreta nitrogen contents of 5.2, 4.6 and 4.7% on a dry matter basis, respectively. This result indicated that the 110% essential amino acid level

was excessive, thus resulting in a reduced apparent nitrogen retention efficiency (63.2, vs. 68.1, 68.9% respectively). Although the current experiment was different from that of Ibrahim (1997), the two experiments indicate that excessive dietary essential amino acids can result in reduced dietary nitrogen retention. From nitrogen retention and excretion point of view, Diet T1 had better amino acid balance and content than Diets T2 and T3 and better protein content than the control diet.

There were significant differences ($p<0.05$) in nitrogen retention efficiency determined in the balance study over the period of 0-7 wk of age (Table 5.3), but there was no significant difference in body protein content among the four bird groups (Table 5.4, $p>0.05$). The possible explanation for this difference is that some body protein was lost during the growing period as feather, scales, downs, etc. Nitrogen retained in the body based on the balance study might have overestimated body protein retention. For example, the protein retained for Diet T1 based on the balance study was about 193 g during 4-6 wk, but the protein deposited in the body in the same period was just 68 g. The body protein loss during the growing period might have elimilated the difference in nitrogen retention measured in the balance study.

CONCLUSION

The three test diets were 1.2-2.2 percentage points lower in dietary protein concentration and contained different essential amino acid profiles as compared with the control diet based on the recommendations of NRC (1994). Lysine, methionine, methionine + cystine, and threonine in Diet T2 and T3 were 6-15% and 11-25% higher than in Diet T1, respectively. The birds fed the three test diets obtained similar growth performance to the control birds. The three low-protein test diets did not increase carcass lipid content. Instead, the relative percentage and absolute amount of body lipid, protein, and breast meat were similar among the four bird groups. Furthermore, the three test diets ($p<0.05$) reduced excreta nitrogen content by 16, 9, and 7% for T1, T2 and T3 as compared with the control diet. The results indicated that the low-protein content with an alternate amino acid balance in Diet T1 with 4-phase feeding not only did not increase body lipids, but also resulted in reduced excreta nitrogen content, similar breast meat yield and growth performance, as compared with the other three diets. Therefore, Diet T1 is

recommended for production use since lower excreta nitrogen concentration and minimal dietary essential amino acids are associated with Diet T1 as compared with Diets T2 and T3.

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CHAPTER VI

EFFECT OF DIETARY CEREALS ON BODY COMPOSITION OF BROILER CHICKENS

ABSTRACT

Wheat and barley have been reported to reduce liver fatty acid synthesis in chickens. Two experiments were conducted to evaluate the effects of barley, wheat, barley hull and wheat bran diets on body lipid deposition in broiler chickens as compared with a corn diet. Exp. 6.1 was a 3 diet × 4 lighting program × 2 sex factorial trial using 2,880 Peterson × Arbor Acres day-old chicks. The three diets, based on corn, barley and wheat, respectively, contained similar protein levels but the barley diet contained about 200 kcal/kg less AME than the corn and wheat diets. Two types of microbial enzymes, mainly containing β-glucanase or xylanase, were added to the barley and wheat diets, respectively. Both the barley and wheat diets reduced bird body lipid accretion by 15.6 and 6.7% respectively by 6 wk of age as compared with the corn diet. Final body weight was not affected. The barley and wheat diets significantly ($p<0.001$) changed the partition of stored energy, with more to lean tissue and less to adipose tissue as compared with the corn diet. Exp. 6.2 tested eight diets based on corn, 50% barley + 50% corn, barley, hull-less barley, 50% wheat + 50% corn, wheat, corn + spent grain, or corn + wheat bran as the main cereal(s), respectively, using 1,600 male Hy-line chicks. Exp. 6.2 showed that all the barley and wheat diets significantly reduced bird fat accretion as compared with the corn diet and indicated that, the higher the percentage of hulled barley or wheat in the diet, the lower the body fat content. It was shown that wheat bran (but not barley spent grain) addition to a corn-based diet reduced body fat deposition compared with the corn diet. It was concluded that chicken body fat deposition could be reduced by dietary inclusion of barley, wheat and wheat bran without affecting final body weight if barley or wheat is used to replace corn in a diet during 3-6 wk of age.

Key words: barley, wheat, spent grains, wheat bran, fiber, body fat, broiler chickens

INTRODUCTION

Dietary cereal, liver lipids and fatty acid synthesis

Dietary cereals have a significant effect on liver lipid synthesis and metabolism. Research on lipid metabolism in avian species fed dietary cereal has been conducted to study the etiology of fatty-liver hemorrhagic syndrome in laying hens. Pearson et al. (1978) compared corn, wheat and barley diets on an iso-energetic and iso-nitrogenous basis. The diets were fed to pullets for 13 wk from point of lay. The number of cases of sub-clinical fatty liver-haemorrhagic syndrome, and the mean total lipid and triacylglycerol contents of the liver decreased in the order of corn, wheat and barley. Jensen et al. (1976) observed that when corn and wheat in various proportions were fed to laying hens, the percent lipid and total lipid per liver increased as the proportion of corn increased. They also found that when fed iso-energetic diets, total liver lipid accumulations were the highest for hens fed grain sorghum, corn or triticale and the lowest for those fed barley, oats or rye. Intermediate levels were obtained with wheat diets made iso-energetic with corn oil or animal fat. Patel et al. (1981) also observed a lower liver lipid content in hens fed a wheat or rye diet than when fed a corn diet. However, Kim et al. (1976) reported that liver fat content was not significantly ($p>0.05$) different between laying hens fed corn and wheat diets. Hens fed the wheat diet contained higher ($p<0.05$) body fat than those fed the corn diet (56 vs. 51% on a dry matter basis).

Some researchers explored the effect of dietary cereals on liver fatty acid synthesis in Japanese quail. Maurice and Jensen (1977) reported that Japanese quail fed a corn-soy diet incorporated more [1-^{14}C]-acetate into liver lipids than fed an iso-energetic and iso-nitrogenous wheat-soy diet. Lipid from livers of birds fed the corn-soy diet was significantly increased in 14:0, 16:0, 16:1 and 18:1 fatty acids and decreased in 18:0, 18:2 and 20:4 fatty acids. This result indicates that the corn fed birds showed a higher proportion of *de novo* synthesized fatty acids in the liver than the wheat fed birds, thus the corn increased liver lipogenesis as compared with the wheat. Maurice and Jensen (1979) confirmed that adult Japanese quail fed a corn-soy diet for 6 wk showed a significantly ($p<0.05$) higher rate of liver fatty acid synthesis measured with [1-^{14}C] acetate as compared with those fed a wheat-soy diet on an iso-energetic, iso-nitrogenous and equi-fat basis.

Reports based on young chickens are very few. Jensen et al. (1976) observed that broiler chicks fed iso-energetic diets containing wheat or corn to 4 wk of age did not differ significantly ($p>0.05$) in liver fat accumulation. However, Chohchi et al. (1984) reported that White Leghorn male chicks fed a barley, rye or wheat diet up to 4 wk of age had significantly ($p<0.05$) lower total lipid (but not triacylglycerol) concentrations in serum and the liver than those fed a corn or hulled rice diet. The liver is the main organ to synthesize fatty acids in the chicken (Leveille, et al., 1975). The reduction of liver lipid and fatty acid synthesis by some cereals compared with corn might lead to a reduction of body fat content in broiler chickens. Therefore, it was hypothesized that it might be possible to replace corn with barley or wheat to reduce body fat deposition in broiler chickens. To date, there have been no such reports published in the literature to the author's knowledge.

Dietary fiber and plasma lipids

The effect of dietary cereals on liver lipid metabolism may be related to their fiber type and characteristics. Numerous studies have shown that soluble fiber sources, such as oat bran, wheat germ, legume seeds, pectins, guar gum, psyllium, as natural food, food fractions or isolated concentrates lower blood total and low-density lipoprotein cholesterol (Cara et al., 1992; Kritchevsky & Story, 1993; Anderson et al., 1994; Welch, 1994; Lairon, 1996). Wheat fiber such as wheat bran, representing insoluble fiber sources exhibited more marked effects on plasma triacylglycerol and triacylglycerol-rich lipoproteins in laboratory animals and humans (Cara et al., 1992; Kritchevsky & Story, 1993; Lairon, 1996). Addition of 10 g oat bran or wheat bran significantly lowered the post-meal serum triacylglycerol increase in humans but rice bran, pea fiber and soybean fiber did not (Cara et al 1992). Decreases in postprandial serum triacylglycerols have also been observed in pigs with test meals supplemented with sugar-beet fiber, wheat bran or guar gums (Lairon, 1996). However, the reports regarding the effect of different dietary fiber sources on liver fatty acid synthesis and body composition in broiler chickens are lacking.

There were two objectives for the current study: (1) to study the effects of barley and wheat diets as compared with a corn diet on body fat content in broiler chickens; (2) to study the effects of different fiber sources (barley bran and wheat bran) on body composition and liver lipid content in growing chickens.

MATERIALS AND METHODS

Experiment 6.1

In Exp. 6.1, the effects of three diets, in conjunction of four lighting programs and sex, on growth performance and body composition of broiler chickens were examined in a 3 × 4 × 2 factorial experiment. The three diets contained corn, barley and wheat as the sole cereal respectively, and were formulated to be iso-energetic and to contain similar levels of protein and amino acids. The barley and wheat diets were supplemented, respectively, with 0.1% Avizyme-SX (mainly β-glucanase) and 0.1% Avizyme-TX (mainly xylanase) supplied by Finfeeds International, UK. The four lighting programs were control light (CON2), Increasing programs 2, 3 and 4 (INC2, INC3 and INC4, respectively) and are reported in details in Chapter VIII.

A total of 2,880 day-old broiler chicks (Peterson × Arbor Acres) was raised in 48 floor pens (either 60 males or females/pen) to 42 d of age. The floor pen seting was the same as in Exp. 5.1 in Chapter V. There were 16 and 24 replicate floor pens for each diet and sex, respectively. Four birds per pen were randomly selected at each of 21 and 42 d of age and were killed by cervical dislocation. The head and neck were then removed, as were wings at the first joint and legs at the hock joint as in Exp. 3.1. The carcasses were weighed, bagged, and frozen at -20 C until later preparation for analysis.

The AME values and nitrogen retention of the three diets (both Starter and Grower) were assayed using the marker method of Scott and Boldaji (1997) as in Exp. 5.1. One hundred and forty-four (144) Arbor Acre × Arbor Acre chicks and 24 Petersime battery cages were used with 6 chicks per cage. Each of the Starter and Grower diets was allocated to four replicate cages. Celite was added to the diets at 1% (w/w) as a marker before fed to the birds. The chicks were raised to 17 d of age and the excreta were collected at 16 d of age.

The frozen chicken carcasses were prepared as in Exp. 3.1 (Chapter III). Acid detergent fiber and neutral detergent fiber contents of feed samples were analyzed using an Ankom Fiber diatomite product, System #200 (Ankom, Fairport, New York). Insoluble ash contents in the diets and the excreta samples were analyzed using the method of Vogtmann et al. (1975). Other laboratory analyses and statistical analyses of all the data were the same as in Exp. 3.1. AME and nitrogen retention were calculated as in Exp. 5.1 in Chapter V.

Table 6.1 Dietary composition and determined nutrient analyses (percentage unless stated) in Exp. 6.1

Ingredients	Corn Diet		Barley Diet		Wheat Diet	
	Starter	Grower	Starter	Grower	Starter	Grower
Corn	56.15	64.02	...	...	...	...
Barley	...	...	56.98	64.76	...	...
Wheat	...	...	...	...	61.58	65.59
Soybean meal	28.59	21.76	26.13	19.14	22.99	20.00
Canola meal	5.00	5.00	5.00	5.00	5.00	5.00
Meat meal	5.00	5.00	5.00	5.00	5.00	4.24
Animal-vegetable fat[1]	2.54	1.48	4.31	3.52	2.81	2.35
Limestone	0.74	0.76	0.79	0.82	0.76	0.89
Vitamin premix[2]	0.50	0.50	0.50	0.50	0.50	0.50
Mineral premix[3]	0.50	0.50	0.50	0.50	0.50	0.50
Dicalcium phosphate	0.37	0.43	0.23	0.26	0.34	0.50
DL-Methionine	0.27	0.22	0.25	0.19	0.26	0.16
Salt (NaCl)	0.24	0.24	0.21	0.21	0.16	0.17
Coxistac	0.10	0.10	0.10	0.10	0.10	0.10
Enzyme[4]	...	...	0.10	0.10	0.10	0.10
Determined analyses (as-fed basis)						
Dry matter	88.3	89.7	88.6	89.1	87.3	88.6
AME, kcal/kg	2996	3032	2836	2744	3020	2963
Crude protein	21.4	21.1	22.2	20.7	21.7	21.4
Crude Fiber	3.4	3.2	5.1	5.2	3.5	3.4
ADF[5]	4.6	4.5	5.9	6.6	4.6	4.4
NDF[5]	10.1	11.2	15.1	16.1	12.1	12.4
Ether extract	4.7	3.7	5.6	4.4	4.5	3.7
Calcium	1.08	0.89	0.96	0.98	1.05	1.00
Phosphorus, total	0.81	0.72	0.72	0.74	0.76	0.73

[1] Tallow: soya oil (1:1) blend.

[2] Supplied per kg diet: vitamin A, 9000 IU; vitamin D_3, 1500 IU; vitamin E, 10 IU; vitamin K_3, 0.5 mg; vitamin B_{12}, 0.007 mg; thiamin, 0.4 mg; riboflavin, 6 mg; folic acid, 1 mg; biotin, 0.15 mg; niacinamide, 35 mg; pyridoxine, 4 mg; choline chloride, 1,000 mg; Ethoxyquin, 0.125 g.

[3] Supplied per kg diet: manganese, 60 mg; copper, 5 mg; zinc, 50 mg; selenium, 0.1 mg; iodine, 0.35 mg.

[4] The barley and the wheat diets were supplemented, respectively, with 0.1% Avizyme-SX (mainly β-glucanase) and 0.1% Avizyme-TX (mainly arabinoxylanase) supplied by Finfeeds International, UK.

[5] ADF: acid detergent fiber; NDF: neutral detergent fiber.

Experiment 6.2

This experiment was a single-factor, completely randomized experimental design with eight diets to study the effect of the fiber portion in barley and wheat on body composition in the

chicken. Hull-less barley, brewer's spent grains, and wheat bran were used in this experiment besides corn, barley, and wheat. A corn diet was again used as the control. Both barley and wheat were used at two levels, 50% and 100% of the grain component, in order to study the quantitative effect of the cereals. Hull-less barley was compared with the regular hulled barley to study the effect of the hull. Brewer's spent grains consist of complete barley less starch that is extracted in the brewing process. The wet spent grains were obtained from Storm Brewing Ltd. in Vancouver, and were sun-dried. The spent grains contained 41.8% neutral detergent fiber on an air-dry basis. Wheat bran, obtained from Prairie Sun Grains, Camrose, Alberta, contained 11.5% crude fiber. The dietary compositions are shown in Table 6.2.

Male commercial Hy-Line chicks (1,600) were obtained at day-old and were fed the eight diets *ad libitum* to 42 d of age in 32 floor pens, 50 chicks each. Each pen measured 4 × 1.5 m. The eight diets were randomly assigned to the 32 pens, four replicate pens for each diet. A 23-h light and 1-h dark regime of lighting was used for all pens during the whole feeding period. Light intensity was 20 lx for 0-7 days and 5 lx for 7-42 days. The other procedures were the same as in Exp. 6.1. Four chicks per pen (i.e., 16 chickens per diet) were randomly collected at each of 3 and 6 wk of age. The chickens were killed by cervical dislocation, the neck artery cut immediately. A 5-ml sample of blood was collected from each bird using a funnel into a test tube with EDTA. After centrifuging, plasma samples were collected and frozen for later analysis.

Dietary AME and nitrogen retention efficiency were measured in a balance study using the total collection method. Two hundred and forty (240) male Hy-line chicks were kept in 36 battery brooders during the first 3 wk, with five chicks per brooder cage and six cages per diet. Feed consumption and excreta output for each cage were accurately measured for three successive days (Sibbald, 1982; NRC, 1994) during the second week. At 3 wk of age, two average-sized healthy birds per cage were moved to individual cages in another room. Feed consumption and excreta output were accurately measured during the fifth week the same way as in the second week. Feed consumption data and excreta outputs of the two birds were averaged as one observation. This provided six observations per diet. The eight diets had different fiber contents, which might affect the transit time of the digesta. After excreta collection in the second and fifth wk, red ferric oxide (Fe_2O_3) was mixed with at 250 mg Fe/kg diet as a colored marker. Feed was removed for 2 h prior to feeding the diets with added Fe_2O_3. A record was made of the

Table 6.2 Dietary composition and determined nutrient analyses (percentage unless stated) in Exp. 6.2

Ingredients	Starter Diets								Grower Diets							
	Control	Barley 50	Barley 100	Hulless 100	Wheat 50	Wheat 100	Spent grains	Wheat bran	Control	Barley 50	Barley 100	Hulless 100	Wheat 50	Wheat 100	Spent grains	Wheat bran
Corn	55.90	27.20	10.00	...	30.40	...	49.30	44.00	64.30	32.00	...	...	33.90	...	56.80	49.60
Barley	...	27.40	44.10	...	...	...	...	...	...	32.30	60.45	...	...	...	...	...
Hulless barley	...	...	...	59.20	...	...	...	...	...	...	...	63.50	...	...	...	...
Wheat	...	...	...	...	30.00	66.00	...	...	...	...	...	...	34.00	72.10	...	...
Spent grains	...	...	...	...	...	...	10.00	...	...	...	...	...	...	...	12.00	...
Wheat bran	...	...	...	...	...	...	...	10.00	...	...	...	...	...	...	...	12.00
Soybean meal	28.80	27.70	27.00	22.70	24.70	19.60	24.10	27.30	25.30	22.20	24.00	22.94	21.90	18.10	19.30	23.90
Corn-soya oil	3.20	5.50	6.80	5.90	2.70	2.10	4.40	6.60	2.10	4.10	7.30	5.70	1.70	1.30	3.40	6.10
Canola meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Meat meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00	...	...	...	...	...	...	...	...
Limestone	0.92	0.95	0.98	0.86	0.94	0.96	0.89	0.93	1.62	1.67	1.69	1.54	1.64	1.66	1.59	1.63
Dicalcium[2]	0.39	0.30	0.21	0.42	0.35	0.31	0.40	0.31	0.94	0.85	0.73	0.95	0.89	0.84	0.95	0.86
DL-Methionine	0.16	0.18	0.18	0.22	0.18	0.20	0.16	0.17	0.07	0.10	0.10	0.13	0.09	0.11	0.06	0.07
L-Lysine HCl	...	...	...	...	0.06	0.17	0.04	...	0.04	1.01	0.03	0.02	0.12	0.21	0.14	0.04
Others[1]	0.65	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.65	0.75	0.75	0.75	0.75	0.75	0.75	0.75
Nutrient Analyses (as fed basis)																
Dry matter	89.2	89.7	89.6	90.1	88.7	89.3	89.3	89.8	88.8	89.0	88.9	89.2	89.4	88.7	88.9	88.7
AME, kcal/kg	2871	2825	2865	2788	2812	2764	2918	2891	3044	2998	2912	2906	2928	2915	3105	3003
Crude protein	21.7	23.3	19.5	20.1	20.2	20.4	20.3	20.2	19.1	19.4	19.4	18.9	19.2	19.5	18.9	18.6
Lysine[4]	1.12	1.12	1.12	1.10	1.1	1.1	1.1	1.12	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Methionine[4]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.38	0.38	0.38	0.38	0.38	0.38	0.38	0.38
Met+Cys[4]	1.2	1.19	1.18	1.139	1.18	1.139	1.19	1.19	1.01	1.19	0.99	0.94	0.99	0.95	1.01	1.01
Tryptaphan[4]	0.28	0.29	0.30	0.28	0.28	0.29	0.28	0.29	0.23	0.29	0.26	0.23	0.23	0.24	0.23	0.24
Threonine[4]	0.79	0.78	0.77	0.80	0.75	0.71	0.81	0.78	0.68	0.78	0.67	0.70	0.64	0.59	0.71	0.67
Crude fat	5.6	5.2	8.0	8.2	5.2	4.1	8.6	8.2	3.2	5.9	7.4	6.9	3.5	2.3	5.8	7.2
Crude fiber[4]	4.0	4.9	5.5	3.8	4.0	3.8	4.9	4.7	3.7	4.8	5.8	3.4	3.7	3.6	4.8	4.6
NDF[3]	10.4	12.4	13.6	13.0	10.4	10.4	13.9	13.0	11.1	13.5	15.5	13.7	11.1	11.0	15.3	14.3
Calcium	0.95	1.1	0.87	0.94	1.0	1.2	0.89	0.92	0.91	0.79	0.84	0.81	0.98	1.10	0.87	1.0
P, available[4]	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.35	0.35	0.35	0.35	0.35	0.35	0.35	0.37

[1] Others included 0.5% vitamin and mineral premix, 0.1% Coxistac (6% premix Brand of Salinomycin Sodium), 0.05% Vitamin E (500,000 IU/kg), 0.1% enzyme except corn diet. The vitamin and mineral premix supplied per kg diet with 120,000 IU vitamin A, 3,000 IU vitamin D_3, 32 IU vitamin E, 3 mg vitamin K, 2.5 mg thiamin, 10 mg riboflavin, 60 mg niacin, 12 mg pantothenic acid, 4 mg pyridoxine, 575 mg choline, 1.2 mg folic acid, 150 mcg biotin, and 18 mcg vitamin B12, 1 mg iodine, 10 mg copper, 60 mg zinc, 75 mg manganese, and 50 mg iron. Enzyme: Bio-Feed Alpha MG, Protease Enzyme, used in barley diets; Bio-Feed plus CT, Endo-1, 4-b-xylanase, Endo-1,4 β-glucanase for wheat, Novo Nordisk A/S, Denmark.

[2] Dicalcium: Dicalcium phosphate ($CaHPO_3$). [3] NDF: neutral detergent fiber. [4] Calculated.

time of feeding and the first appearance of red excreta on the tray under the cage. The time difference was calculated as the transit time.

The frozen carcasses were put into individual autoclave bags and cooked in a small laboratory autoclave oven at 124 C over-night. The subsequent treatment and analyses of the carcass samples were the same as in Exp. 6.1. Gross energy, crude protein, crude fat, neutral detergent fiber, and moisture of the feed and excreta samples were analyzed as in Exp. 6.1. The frozen plasma samples were thawed. Plasma very low-density lipoprotein (VLDL) concentration was analyzed using the method of Griffin and Whitehead (1982) to associate with liver lipid and body fat content since plasma VLDL was reported to have good correlation with bird body fat content (Griffin and Whitehead, 1982).

All the data were analyzed for the main effect of each factor (diet, lighting program and sex) and their interactions by analysis of variance using SAS® (SAS Institute Inc., Cary, NC, 1996).

RESULTS

Experiment 6.1

Growth performance

All three diets were similar in dietary nutrient content except that the barley diet had 200 kcal/kg less AME, higher dietary fat and fiber (neutral detergent fiber and acid detergent fiber) than the corn and wheat diets.

The birds fed the barley diet attained similar final body weight to the control birds. The birds fed the wheat diet attained higher ($p<0.05$) final body weight than the control birds at 6 wk of age, although the chicks fed the wheat diet had lower body weight than those fed the corn and barley diets at 3 wk of age ($p<0.01$). Male birds had higher body weight than female birds at both 3 and 6 wk of age ($p<0.001$, Table 6.3). Male birds fed the wheat diet had lower body weight than males fed the barley diet at 3 wk of age (635 vs 670 g/bird, $p<0.01$, not listed in the table). However, female birds fed the two diets had similar 3-wk body weight, i.e., 610 vs. 617 g/bird for the wheat and barley diets, respectively.

The chicks fed the barley diet had higher feed intake but lower AME intake during 3-6 and 0-6 wk ($p<0.05$) as compared with those fed the corn and wheat diets. Males always had higher intake of feed, AME, and protein than females ($p<0.001$, Table 6.3). Feed: gain ratio was higher with the barley diet during both 0-3 and 3-6 wk and higher with the wheat diet during 0-3 wk as compared with the corn diet ($p<0.05$, Table 6.3). Male birds had lower feed: gain ratio than female birds at all times ($p<0.001$).

AME/gain ratio was in the order of the wheat > corn > barley diets during 0-3 wk ($p<0.01$, Table 6.4) and in the order of the corn > wheat or barley diets during 3-6 wk ($p<0.05$, Table 6.4). The protein/gain ratio was higher with the barley diet than with the corn diet during 0-3 ($p<0.01$) and 3-6 wk ($p<0.05$), with the wheat diet intermediate. Male birds used less dietary energy and more protein for a kg body weight gain than female birds ($p<0.001$ Table 6.4). The nitrogen retention efficiencies for both the starter and the grower phases was significantly higher ($p<0.05$) for the wheat diet than for the corn and barley diets which did not differ ($p>0.05$, Table 6.4).

Body composition

The chicken body fat content at 6 wk was significantly ($p<0.01$) lower for the birds fed the barley and wheat diets as compared with the birds fed the corn diet (12.4, 13.7 vs. 14.7%, respectively, Table 6.5). Even at 3 wk of age, birds fed the barley diet had a significantly lower carcass fat content than the wheat or corn fed birds (8.0 vs. 8.9 or 9.2% respectively, $p<0.05$, Table 6.5). The body protein content in the barley fed birds at 6 wk was significantly higher than that in the corn or wheat fed birds (19.3 vs. 18.9 or 18.9% respectively, $p<0.01$, Table 6.5). The carcass energy content differences among the diets had the same pattern as the carcass fat content at 6 wk, i.e., the barley < wheat < corn fed birds ($p<0.05$). However, under the Increasing lighting program 3, the carcass energy content pattern was different, i.e., the wheat < barley, or corn fed birds (2215 < 2321 or 2349 kcal/g, respectively, $p<0.05$, not listed in the tables). The carcass contained, on average, 71% and 65% water at 3 and 6 wk, respectively.

Males consistently had higher carcass water and lower carcass fat and energy contents than females at both 3 and 6 wk of age ($p<0.001$). Male birds contained higher carcass protein ($p<0.001$) at 6 wk but similar protein at 3 wk as compared with female birds.

Energy or protein retention efficiency was defined as energy or protein retained in the body/AME or protein intake, respectively. Birds fed the three diets had similar energy retention and protein retention efficiencies during 0-3 and 0-6 wk (Table 6.5, $p>0.05$). Males also had similar energy retention and protein retention efficiencies to females during both 0-3 and 3-6 wk of age.

Table 6.5 also shows that the proportion (k_f) of energy retained as fat/the total retained energy during 0-6 wk was in the order: k_f corn > k_f wheat > k_f barley diet (i.e., 0.56 > 0.54 > 0.51, respectively), with each significantly different from the other ($p<0.001$). This means that a higher proportion of retained energy went to body fat in the corn fed birds than in the barley and wheat fed birds. During 0-3 wk, k_f corn and k_f wheat > k_f barley diet (i.e., 0.45, 0.45 > 0.42, respectively, $p<0.001$, Table 6.5). Under control lighting 2, the k_f for birds fed the three diets during 0-3 wk were similar (0.46, 0.47 and 0.46, respectively, for the corn, wheat and barley

Table 6.3 Effect of diets and sex on the growth performance in Exp. 6.1

	Body weight, g		Feed: gain ratio, g/g			Feed intake, g/d/bird			AME intake, kcal/d/bird			Protein Intake, g/d/bird		
	3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk
Diet														
Corn	641[a]	2144[b]	1.41[b]	1.84[b]	2.04[b]	40	145[b]	90[b]	121[a]	440[a]	280[a]	8.6[b]	30.6[b]	19.6[b]
Barley	639[a]	2148[b]	1.44[a]	1.92[a]	2.13[a]	41	153[a]	95[a]	116[b]	419[b]	267[b]	9.1[a]	31.7[a]	20.3[a]
Wheat	626[b]	2174[a]	1.45[a]	1.86[b]	2.02[b]	40	148[ab]	92[ab]	122[a]	440[a]	281[a]	8.8[b]	31.7[a]	20.3[a]
SEM[1]	2.8	8.4	0.01	0.01	0.01	0.001	0.001	0.001	0.9	2.8	1.4	0.07	0.2	0.1
Significance	**	*	*	*	*		*	*	**	**	**	**	**	**
N[1]	440	430	16	16	16	16	16	16	16	16	16	16	16	16
Sex														
Male	657[a]	2356[a]	1.40[b]	1.83[b]	2.00[b]	41	161[a]	99[a]	122[a]	469[a]	295[a]	9.0[a]	34.0[a]	21.5[a]
Female	615[b]	1957[b]	1.46[a]	1.92[a]	2.13[a]	40	136[b]	86[b]	118[b]	396[b]	257[b]	8.7[b]	28.7[b]	18.7[b]
SEM	2.3	6.9	0.01	0.01	0.01	0.001	0.001	0.001	0.7	2.3	1.15	0.05	0.17	0.08
Significance	***	***	***	***	***		***	***	***	***	***	***	***	***
N	660	645	24	24	24	24	24	24	24	24	24	24	24	24
Interaction[2]														
Diet × sex	***													

[abcd] Means with a different letter are significantly different, * $p \leq 0.05$, ** $p \leq 0.01$, *** $p \leq 0.001$.

[1] SEM: standard error of mean; N: number of observations.

[2] The significant ($p<0.05$) interaction between light programs and sex is listed in Table 8.4, Chapter VIII.

Table 6.4 Effect of diet and sex[1] on nitrogen retention, AME/gain and protein/gain in Exp. 6.1

	AME/gain, kcal/kg[3]			Protein/gain, g/kg[3]			Nitrogen Retention[2]	
	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk
Diet								
Corn	4208^{b}	6176^{a}	5573 ^{a}	301^{b}	430^{b}	390^{b}	0.39^{b}	0.39^{b}
Barley	4076^{c}	5857^{b}	5349 ^{b}	319^{a}	442^{a}	405^{a}	0.39^{b}	0.39^{b}
Wheat	4391^{a}	6000^{b}	5523 ^{a}	315^{a}	434^{ab}	398^{ab}	0.43^{a}	0.44^{a}
SEM[4]	19	42	30	1	3	2	0.01	0.01
Significance	**	*	**	**	*	**	*	*
N[4]	16	16	16	16	16	16	4	4
Sex								
Male	4146^{b}	5824^{b}	5343 ^{b}	318^{a}	449^{a}	407^{a}		
Female	4305^{a}	6199^{a}	5600 ^{a}	306^{b}	421^{b}	388^{b}		
SEM	16	34	24	1	3	2		
Significance	***	***	***	***	***	***		
N	24	24	24	24	24	24		

^{abc} Means with different superscripts are significantly different, * p≤0.05; ** p≤0.01, ***: p≤0.001.

[1] Interaction between diet × sex for the above parameters was not significant (p>0.05).

[2] Nitrogen retention was measured in 16 d old chicks in a separate trial.

[3] AME/gain = feed/gain × dietary AME content (kcal/kg); Protein/gain (g/kg) = feed/gain × dietary protein (%) × 10.

[4] SEM: standard error of mean; N: number of observations.

diets, not listed in the tables). Male birds partitioned less retained energy to body fat (*kf*: 0.52 vs. 0.56, respectively, during 0-6 wk, p<0.001, Table 6.5) and more energy to body protein than female birds.

Experiment 6.2

Dietary nutrient contents

The hull-less 100 and wheat 100 diets fed during the first 3 wk had about 100 kcal/kg lower AME than the control diet (Table 6.2). The barley 100, hull-less 100 and wheat 50 diets fed during 3-6 wk also had around 100 kcal/kg lower AME than the control diet and the spent grain diet. Otherwise, the eight diets had similar energy and protein contents. The three barley diets and the spent grain diet were supplemented with protease by mistake, rather than β-glucanase

Table 6.5 Effect of diets and sex on carcass composition and energy retention in Exp. 6.1

	Carcass water[1], %		Carcass protein[1], %		Carcass fat[1], %		Carcass energy[1]	Protein retention[2]		Energy retention[2]		k_f[3]	
	3 wk	6 wk	3 wk	6 wk	3 wk	6 wk	at 6 wk	0-3 wk	0-6 wk	0-3 wk	0- 6 wk	0-3 wk	0-6 wk
Diet													
Corn	70.4	63.9 ^{c}	18.2	18.9^{b}	9.2 ^{a}	14.7 ^{a}	2433^{a}	0.59	0.48	0.43	0.42	0.45^{a}	0.56^{a}
Barley	70.9	65.8 ^{a}	18.2	19.3^{a}	8.0 ^{b}	12.4 ^{c}	2239^{c}	0.58	0.48	0.43	0.42	0.42^{b}	0.51^{c}
Wheat	70.9	64.9 ^{b}	18.2	18.9^{b}	8.9 ^{a}	13.7 ^{b}	2331^{b}	0.58	0.48	0.43	0.42	0.45^{a}	0.54^{b}
SEM[4]	0.2	0.3	0.1	0.1	0.1	0.2	28	0.003	0.003	0.003	0.003	0.00	0.00
Significance		*		**	**	**	*					***	***
N[4]	63	63	63	63	63	63	63	16	16	16	16	63	63
Sex													
Male	71.5 ^{a}	65.8 ^{a}	18.2	19.1^{a}	8.1 ^{b}	12.5 ^{b}	2242^{b}	0.59	0.48	0.43	0.43	0.42^{b}	0.52^{b}
Female	70.0 ^{b}	63.9 ^{b}	18.1	18.9^{b}	9.3 ^{a}	14.8 ^{a}	2426^{a}	0.58	0.48	0.43	0.42	0.45^{a}	0.56^{a}
SEM	0.2	0.3	0.1	0.1	0.1	0.2	23	0.002	0.002	0.002	0.002	0.00	0.00
Significance	***	***		***	***	***	***					***	***
N	95	95	95	95	95	95	95	24	24	24	24	95	95
Interaction[5]													
Diet × light							*					*	

[abc] Means with different superscripts are significantly different, * p≤0.05; ** p≤0.01, ***: p≤0.001.

[1] On fresh weight basis. Carcass energy (kcal/g) was measured using bomb calorimeter. Carcass protein (%) = 100 - water % - fat % - ash %.

[2] Energy retention efficiency = energy retained in bird/AME consumed ≈ average carcass energy/g of four birds from a pen/(AME/gain). Protein retention efficiency = [average carcass protein % of the four birds from a pen × average body weight (g)]/protein intake (g).

[3] Partition coefficient of stored energy to fat (k_f)= energy stored as fat/total stored energy.

[4] SEM: standard error of mean; N: number of observations.

[5] Only significant interactions are listed. The effects of lighting programs were reported in Table 8.5 in Chapter VIII.

(see Table 6.2). Generally, the two barley diets, spent grain diet, and wheat bran diet had higher fat and neutral detergent fiber contents than the two wheat and corn diets.

The three barley diets (barley 50, barley 100 and hull-less 100) had relative shorter transit times (155-160 min) compared with the corn and wheat diets (164-173 min) at 16 d of age. The three barley diets transited faster than the corn diet (202-204 vs. 225 min, respectively) and transited more slowly than the three wheat diets (182-191 min) on Day 35. The older the birds, the longer the feed passage time. The correlation between dietary neutral detergent fiber and the passage time was low and inconsistent ($r = -0.40$ for 16 d and 0.19 for 35 d of age, respectively).

Growth performance

By 6 wk of age, all the bird groups obtained similar body weight except that the birds fed barly 100 and hull-less barley 100 diets had significantly lower ($p<0.05$) body weight than the control birds. At 3 wk of age, the birds fed the seven test diets had significantly lower body weights than the corn diet ($p<0.05$, Table 6.6). The higher the barley or wheat percentage in a diet, the lower the chick body weight (Table 6.6). All the birds fed the test diets had similar feed, AME and protein intake to the control birds, except that the birds fed the barley 100, hull-less 100 and wheat 50 diets had lower ($p<0.05$) AME intake than those fed the control diet during 0-6 wk.

The birds fed the seven test diets used more feed (Table 6.6), AME and protein (Table 6.7) for a unit body weight gain than the control birds during 0-3 wk. However, during 3-6 wk, the pattern was reversed. The ratios of feed/gain, AME/gain and protein/gain were significantly ($p<0.05$) or non-significantly lower in the birds fed the test diets (but the spent grain diet) than in the corn fed birds.

Body composition

The body fat at 3 wk was basically not affected by the test diets. However, The body fat content at 6 wk of age was significantly lower in the birds fed all the test diets but wheat 50 and spent grain diets ($p<0.01$, Table 6.7) than in the birds fed the corn diet. The higher the barley or wheat percentage in a diet, the lower the body fat content. The barley diets more significantly reduced body fat than did the wheat diets. The birds fed the test diets, except wheat 50 and the spent grain diets, were 14.3 - 21.9% lower in body fat content at 6 wk of age than the control.

Table 6.6 Effect of diets on bird performance and AME and protein intake (Exp. 6.2)

Diet	Body weight, g		Feed intake, g/d/bird			AME intake, kcal/d/bird			Protein intake, g/d/bird			Feed: gain ratio, g/g		
	at 3 wk	at 6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk
Control	746^{a}	2523^{a}	44	161^{ab}	98^{ab}	126^{ab}	487^{ab}	290^{b}	9.5^{ab}	30.8^{ab}	19.1^{ab}	1.33^{c}	1.90^{ab}	1.72^{ab}
Barley 50	678^{cd}	2484^{ab}	43	158^{ab}	97^{b}	122^{b}	474^{bc}	286^{bc}	10.1^{a}	30.7^{ab}	19.3^{a}	1.45^{ab}	1.83^{ab}	1.72^{ab}
Barley 100	669^{d}	2461^{b}	44	155^{ab}	96^{b}	127^{ab}	450^{c}	279^{c}	8.7^{c}	30.0^{ab}	18.9^{ab}	1.50^{a}	1.82^{ab}	1.73^{ab}
Hull-less 100	673^{d}	2451^{b}	44	159^{ab}	96^{b}	123^{b}	462^{bc}	278^{c}	8.9^{bc}	30.0^{ab}	17.5^{ab}	1.52^{a}	1.84^{ab}	1.76^{ab}
Wheat 50	724^{b}	2504^{ab}	44	153^{b}	95^{b}	125^{b}	449^{c}	276^{c}	9.0^{bc}	29.4^{ab}	18.3^{ab}	1.39^{bc}	1.81^{b}	1.68^{b}
Wheat 100	727^{b}	2487^{ab}	45	155^{ab}	96^{b}	125^{b}	453^{c}	276^{b}	9.2^{bc}	30.3^{ab}	18.5^{ab}	1.41^{b}	1.86^{ab}	1.72^{ab}
Spent grain	694^{c}	2486^{ab}	46	163^{a}	100^{a}	134^{a}	507^{a}	307^{a}	9.3^{bc}	31.2^{a}	19.7^{a}	1.46^{ab}	1.91^{a}	1.80^{a}
Wheat bran	714^{b}	2480^{ab}	44	156^{ab}	97^{b}	127^{ab}	468^{bc}	287^{bc}	8.9^{bc}	29.0^{b}	18.0^{b}	1.39^{bc}	1.86^{ab}	1.72^{ab}
SEM[1]	7	23	1	2	1	2	7	3	0.1	0.4	0.2	0.02	0.03	0.02
Significance	*	*		*	*	*	*	*	**	*	**	*	*	*
N[2]	100	98	4	4	4	4	4	4	4	4	4	4	4	4

[abcd] Means with different superscripts are significantly different, * $p<0.05$; ** $p<0.01$.

[1] Standard error of mean.

[2] Number of observation.

Table 6.7 Effect of diets on AME/gain, body fat content, and serum VLDL concentrations (Exp.6.2)

Diet	AME/gain, kcal/kg			Protein/gain, g/kg			Body lipids[1], %		Liver lipids[1], %	Serum VLDL[2]	
	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	3 wk	6 wk	6 wk	3 wk	6 wk
Control	3809^{c}	5804^{b}	5212^{b}	289^{bc}	366^{a}	342^{ab}	9.3^{ab}	12.6^{a}	9.4^{a}	0.06^{b}	0.25
Barley 50	4027^{b}	5492^{c}	5067^{bc}	337^{a}	355^{ab}	350^{a}	9.0^{b}	10.5^{b}	8.4^{ab}	0.08^{ab}	0.26
Barley 100	4300^{a}	5294^{d}	5014^{bc}	293^{bc}	353^{ab}	336^{ab}	9.7^{ab}	9.4^{b}	7.2^{b}	0.06^{b}	0.22
Hull-less 100	4241^{a}	5477^{c}	5105^{bc}	306^{b}	348^{b}	337^{ab}	9.9^{ab}	10.5^{b}	8.0^{ab}	0.07^{b}	0.22
Wheat 50	3897^{bc}	5304^{d}	4878^{c}	280^{c}	348^{b}	327^{b}	10.9^{a}	11.4^{ab}	7.7^{ab}	0.10^{ab}	0.27
Wheat 100	3909^{bc}	5412^{cd}	4948^{c}	289^{bc}	362^{ab}	339^{ab}	10.3^{ab}	10.3^{b}	7.8^{ab}	0.15^{a}	0.31
Spent grain	4249^{a}	6015^{a}	5478^{a}	296^{bc}	366^{a}	347^{a}	10.6^{ab}	12.9^{a}	8.3^{ab}	0.07^{b}	0.22
Wheat bran	4025^{b}	5577^{c}	5114^{bc}	281^{bc}	345^{b}	326^{b}	9.9^{ab}	9.7^{b}	7.7^{ab}	0.09^{ab}	0.26
SEM[3]	41	18	55	4	5	4	0.4	0.4	0.6	0.02	0.05
Significance	*	**	*	**	*	*	*	**	*	*	
N[3]	4	4	4	4	4	4	13	16	16	14	14

^{abcd} Means with different superscripts are significantly different, * $p<0.05$; ** $p<0.01$.

[1] On a fresh basis.

[2] VLDL: very low-density lipoprotein measured based on turbidity produced by precipitation of VLDL using 0.25 g heparin/L (150 mM $MgCl_2$, 154 mM NaCl, 10 mM Tris-HCl) solution and measured at 540 nm (Griffin and Whitehead, 1982).

[3] SEM: standard error of mean; N: number of observations.

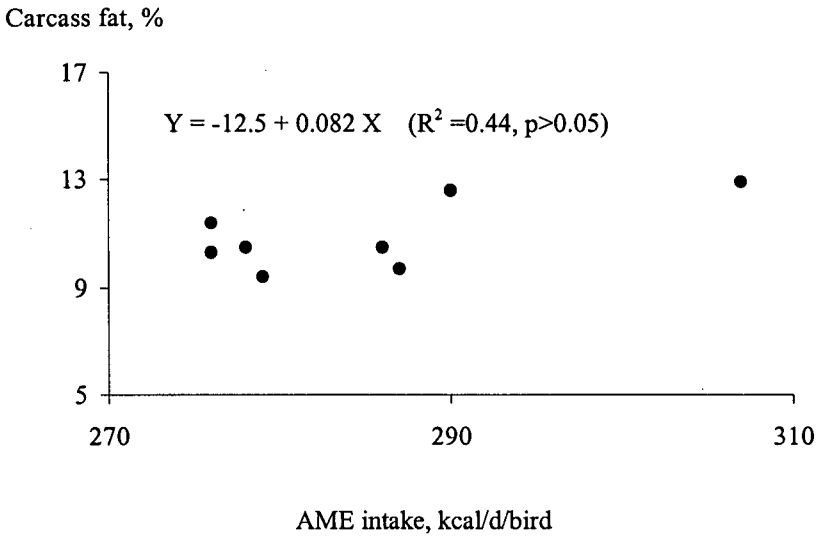

Fig. 6.1 Relationship between body lipid content and AME intake (Exp. 6.2)

Liver lipid content in birds fed the barley 100 diet at 6 wk was significantly lower than in those fed the corn diet ($p<0.05$, Table 6.7). The liver lipid contents in birds fed the other diets were intermediate and were not significantly different ($p>0.05$). The liver lipid contents had very consistent change with the body fat content (Table 6.7). The correlation coefficient between liver lipid and body fat contents was 0.74 ($p<0.01$). Corn-fed chickens at 6 wk also had higher liver weight (70.0 g) than the liver (60.6-62.4 g) from other bird groups.

The body lipid content at 6 wk was plotted againsty AME intake (Fig. 6.1). The linear regression relationship was not significant ($p>0.05$). The body lipid content at 6 wk was highly correlated with AME/gain ratio ($r = 0.77$, $p<0.05$, Table 6.7). It means that the higher the body lipid content, the higher the energy amount required per unit weight gain (i.e., AME/gain ratio).

Serum VLDL

The chickens fed the wheat diets tended to have higher VLDL at both 3 and 6 wk of age than the other chicken groups, but only significantly higher ($p<0.05$, Table 6.7) than those fed the corn, barley 100 and hull-less 100 diets at 3 wk of age. The plasma VLDL of the chickens at 6 wk was three times that at 3 wk. The VLDL concentrations were found to be poorly correlated with bird body fat content, with correlation coefficients of 0.37 and -0.20 at 3 and 6 wk, respectively (Table 6.7).

DISCUSSION

Experiment 6.1

Growth Performance

The birds fed the barley diet with 200 kcal/kg lower dietary energy and the wheat diet achieved similar or even higher final body weights than the corn fed birds, although the birds fed the wheat diet had lower 3-wk body weight than the corn and the barley fed chicks.

Birds fed a barley diet are expected to have lower body weight than corn fed birds due to the β-glucans in barley (Annison and Choct, 1991). It is interesting to speculate why the birds fed the barley diet achieved similar 3-wk and 6-wk body weight at lower energy intake when compared with the corn-fed birds. The possible explanation is the reduced body fat content in the barley-fed chicks as compared with the corn-fed chicks (8.0 vs. 9.2% at 3 wk, 12.4 vs. 14.7%

at 6 wk, $p<0.01$, Table 6.5). Since the barley-fed chicks deposited less fat in the body, more of the ME intake was used to synthesize lean tissue. The energy required to deposit 1 g of fat could be repartitioned to deposit 2.7 g of lean tissue, thus increasing body weight at a given amount of ME intake when less fat and more lean tissue is deposited. The reduced body fat deposition may have reversed the growth depressing effect of the lower ME intake and β-glucans existing in the barley diet. The resultant effect was a similar 3- and 6-wk body weight in the chicks fed the barley and the corn diets.

The birds fed the wheat diet had significantly lower body weight (626 vs. 641 g/bird, $p<0.01$) at 3 wk but higher body weight (2174 vs. 2144 g, $p<0.05$) at 6 wk than the control birds (Table 6.3). It is probable that the anti-nutritive factors (e.g., arabinoxylans), to some extent, still present in this diet supplemented with Avizyme TX, affected the early growth of the chicks fed the wheat diet (Annison and Choct, 1991). The wheat diet did not reduce chick body fat content as the barley diet did, therefore, the body weight of the wheat fed chicks did not catch up with the corn fed birds as the barley fed birds did, as explained above. At 6 wk of age, the wheat diet reduced body fat accretion, thus affecting body weight as explained for the barley diet above.

The wheat diet reduced 3-wk body weight of males but not of females compared with the barley diet. This result suggests that the anti-nutrients present in wheat had a greater effect on male chicks than on female chicks.

As to the lower efficiency of feed conversion to body weight with the barley and the wheat diets during 0-3 wk, it is likely accounted for by the growth-depressing effects of β-glucans mainly in barley and arabinoxylans mainly in wheat and the lower AME content in the barley diet. Microbial enzymes (β-glucanase and xylanase) which hydrolize the non-starch polysaccharides can partially overcome, but cannot completely elimilate the anti-nutritive effect, especially in young birds (Annison and Choct, 1991). However, the wheat diet had a similar feed/gain ratio to the corn diet during 3-6 wk (Table 6.3). This result suggested that the growth-restraining effects of the anti-nutrients in the wheat are largely alleviated in the older chickens. The barley diet had a significantly higher feed/gain ratio during 3-6 wk than the corn diet (1.92 vs. 1.84, $p<0.05$), due probably to a lower AME content in the barley diet.

Feed/gain, AME/gain and protein/gain ratios reflect the efficiencies of dietary metabolizable energy and protein utilized for body weight gain. Obviously, the change in body

composition and existence of dietary growth depressing factors significantly affected these ratios. The possible explanation is similar as for the body weight differences above.

Body composition, energy retention and partition

The barley fed birds had a significantly lower carcass fat content than the corn fed birds at both 3 and 6 wk (8.0 vs. 9.2% at 3 wk, 12.4 vs. 14.7% at 6 wk, $p<0.05$, Table 6.5). The wheat fed chickens also had a 6.7% lower body fat content at 6 wk (but not at 3 wk) than the control chickens.

The reduction of body fat by 15.6% at 6 wk with the barley diet was comparable to that obtained with genetic selection. Whitehead (1988) reported an 18.3% reduction in body lipid content through seven generations of successive genetic selection for lean birds. The body fat was reduced by the barley and wheat diets without reducing final body weight (Table 6.3). The feed cost in the current study was also reduced due to the lower prices of barley and wheat compared with corn.

It is affirmative that the lower dietary energy content in the barley diet should be part of the explanation for the lower body fat content in the barley fed birds, but not the complete explanation. Generally, a reduction in dietary energy results in lower body weight in broilers (Jackson et al., 1982). However, the body weight of the barley fed birds at lower energy intake was similar to that of the corn fed birds. This could be true only when body fat content of the barley fed birds was reduced, and more of the retained energy was used for lean tissue deposition as a gram of lean meat deposition needs less energy than a gram of adipose tissue does. The lower AME requirement for a unit weight gain for birds fed the barley diet compared with the corn diet (5349 vs. 5573 kcal/kg, $p<0.001$, Table 6.4) supports the above explanation.

Table 6.5 shows that a lower proportion (k_f) of retained energy was directed to body lipid deposition for the barley fed birds than for the corn fed birds (0.51 vs. 0.56, $p<0.001$). The lower body fat content in the barley fed birds was probably because the barley diet changed the partition of retained energy between body fat and protein deposition, with more to body protein and less to body fat. Likewise, the k_f for the wheat diet (0.54) was significantly lower than the k_f (0.56) for the corn diets ($p<0.01$, Table 6.5). The same explanation applies. The barley diet more greatly redeced body fat deposition than the wheat diet, which was indicated by the lower

6-wk body fat content ($p<0.01$) and lower k_f ($p<0.001$) for the barley fed birds than for the wheat fed birds.

The similar dietary energy and protein retention efficiencies among the diets indicate that the diets did not significantly ($p>0.05$) change dietary energy and protein utilization for body energy and protein depositon. The wheat diet (both starter and grower formulae) gave significantly ($p<0.05$) higher nitrogen retention efficiency than the corresponding corn and barley diets (0.44 vs. 0.39 and 0.39). This result, together with higher liver lipids and final body weight associated with the wheat diet, indicates that wheat may have some special nutritional characteristics.

Sex differences in performance and body composition

Males had a higher feed intake, higher body weight, and lower feed/gain ratio than females ($p<0.001$, Table 6.3). Male birds contained less fat and more protein and water in their bodies, and thus had lower energy content per unit weight than female birds (Table 6.5). Males partitioned less retained energy to body fat and more to body protein deposition ($p<0.001$, Table 6.5) than females. These effects may be related to genetic and endocrine differences.

The diets had significant interaction with the lighting programs in carcass energy content. Under Increasing lighitng program 3 (INC3), barley fed birds contained higher energy ($p<0.05$) than wheat fed birds at 6 wk. It could be that INC3 resulted in the non-significantly higher adipose tissue compensatory growth for the barley fed birds than for the wheat fed birds at a late age, thus leading to higher carcass energy in the barley fed birds than the wheat fed birds raised under INC3.

Experiment 6.2

Supplementing the three barley diets and spent grain diet with protease rather than β-glucanase may explain why the barley diets had lower AME contents than the corn diet. The corn diet had the longest passage time among the eight diets. The barley diet had the shortest transit time in the second wk, while the wheat diet transited most quickly in the fifth wk. The longer the passage time, the more complete the feed digestion. This may partially explain why the barley diet had lower nutrient digestibility. The correlation between dietary neutral detergent

fiber and the passage time was low and inconsistent (r = -0.40 for Day 16 and 0.19 for Day 35). Obviously, neutral detergent fiber is not a good parameter reflecting the feed transit speed. The passage time is related to the type of fiber components and the cereal particle size (Lupton et al., 1993). In human volunteer studies, both barley bran (Lupton et al., 1993) and whole wheat meal (Wiggins et al., 1986) accelerated transit of food. The current results support these findings.

Growth performance

The body weight at 3 wk was in the order of birds fed the corn > wheat, wheat bran > barley, hull-less barley 100 and barley spent grain diets. These results reflect the growth-depressing effect of the non-starch polysaccharides in barley and wheat, more seriously in barley, as discussed in Exp. 6.1, and their lower dietary AME contents and intakes. Different from that in Exp. 6.1, the body weight of the barley fed chicks at 3 wk was lower than that of the wheat fed birds. This was probably due to differences in experimental conditions between experiments.

The birds fed all seven test diets, to different extents, caught up with the corn fed birds in body weight at 6 wk, due to the alleviation of the growth depressing effect of the anti-nutrients in the barley and wheat. It can be concluded that the effect of barley and wheat on growth is more severe at an early age than at later ages and is still somewhat evident during 3-6 wk depending on barley or wheat type (Scott, et al., 1998) and inclusion percentage in the diet.

As in Exp. 6.1, the birds fed test diets except wheat 50, wheat 100 and wheat bran diets had significantly ($p<0.05$) higher ratios of feed/gain (Table 6.6) and AME/gain (Table 6.7) during 0-3 wk than the control birds. During 3-6 wk, however, the control birds had higher ($p<0.05$) AME/gain ratio than birds fed the test diets except spend grain diet (Table 6.7). These results probably resulted from the effect of anti-nutrients in the test diets during 0-3 wk and the dramatic change in body composition of the birds fed the test diets (but the spent grain diet) during 3-6 wk, which was discussed in Exp. 6.1.

Body composition

Consistent with the results in Exp. 6.1, the final body fat content was significantly ($p<0.01$) reduced in birds fed the three barley diets and the wheat 100 diet as compared to birds fed the corn diet. The higher the barley or wheat percentage in a diet, the lower the body fat content.

These results indicate a quantitative relationship between the cereal percentage in a diet and body fat content. The barley diets more significantly reduced the bird's body fat than the wheat diets, consistent with the results in Exp. 6.1. In Exp. 6.2, the test diets, except the spent grain diet, reduced bird body fat by 14.3 - 21.9% as compared with the corn diet. The fat-reducing effect was surprising and was as great as that produced by feeding broilers a high-protein diet (Kirchgenesser et al., 1978; Jackson et al., 1982).

Based on the non-significant ($p>0.05$) linear regression relationship between body fat content and AME intake during 0-6 wk, it can be speculated that the difference in dietary AME intake is not the main explanation for the difference of body fat content in birds fed the eight diets. The main reason is probably that the barley and the wheat diets altered the partition of retained energy, with more to protein and less to fat deposition, as shown in Exp. 6.1 (Table 6.2).

The liver lipid content showed a very consistent change with the body fat content (Table 6.7). The correlation coefficient between liver lipid and body fat contents was 0.74. This result suggests that body fat content differences among the diets were due to differences in rates of liver lipid synthesis. In this experiment, the corn-fed birds had higher liver lipid content than the barley, wheat or wheat bran fed birds. This result strongly supports that of Jensen et al. (1976) and Maurice and Jensen (1979) who observed that corn fed laying hens or Japanese quail had higher liver fat content and rate of heptic fatty acid biosynthesis than wheat-fed birds. The corn-fed chickens at 6 wk also had higher liver weight (70.0 g) than the other bird groups (60.6-62.4 g) in the current study.

The body fat content and AME/gain are two independently measured parameters. The body fat content at 6 wk was highly correlated with the AME/gain ratio ($r = 0.77$). The higher the body fat content, the higher the amount of energy required per unit weight gain (i.e., AME/gain ratio, Table 6.7). This is because the requirement of dietary energy for a unit of weight gain decreased when carcass fat and energy content decreased as shown in Exp. 6.1.

The wheat bran diet containing 12% wheat bran gave a significant ($p<0.01$) reduction in body fat content as compared with the corn diet. The wheat 100 diet for the grower period included 72% wheat. Since wheat contains about 20% bran, the wheat 100 diet contained approximately 14% wheat bran, which is close to the 12% wheat bran inclusion level in the wheat bran diet. The two diets had a similar fat-reducing effect therefore, presumably, the fat-reducing factor(s) of wheat are mainly in the bran fraction.

The spent grain diet did not result in a reduction in body lipids as compared with the corn diet. This suggests that the external layer of barley does not have a lipid-reducing factor(s) provided the processing procedure of the spent grains did not change the nature of the external layer of barley. The hull-less 100 diet used in the current study was intended to compare with the barley 100 diet to study the effect of the hull. Both the hull-less 100 and barley 100 diets had similar effects on body lipid content. Therefore, it seems that the barley hull did not affect body lipid content either.

Plasma VLDL concentration

The plasma VLDL concentrations did not show consistent changes with body fat and liver lipid content. The poor correlation between VLDL concentrations and bird body fat content indicates that plasma VLDL concentration does not reflect the fatness of birds. However, Griffin and Whitehead (1982) reported a correlation of 0.70 and 0.65, respectively, between plasma VLDL concentration and body lipid content for males and females. One explanation for the difference may be that Griffin and Whitehead (1982) took duplicate blood samples for each bird, 2 d apart. Only the mean of two consistent plasma VLDL levels was used to plot against body lipid content to calculate the correlation coefficient in their study. In the current study, only one blood sample was taken per bird. Griffin et al. (1982) also observed a low correlation of 0.3-0.5 between single measurement of plasma VLDL and body lipid content. Chickens fed the wheat diets tended to have higher VLDL at both 3 and 6 wk of age than other chicken groups. Higher concentrations of total and VLDL + LDL cholesterol were found in rats fed wheat bran with cholesterol than in those fed oat bran, barley or malted barley with cholesterol (Jackson et al., 1994). A higher VLDL concentration in the wheat fed chickens than in the barley or corn fed birds has not yet been reported.

CONCLUSION

It was found that the inclusion of barley, wheat and wheat bran in broiler diets significantly ($p<0.05$ or 0.01) reduced body fat content without significantly ($p>0.05$) affecting final body weight. The higher the barley or wheat percentage in a diet, the lower the body fat content. The barley and the wheat diets significantly ($p<0.001$) altered the partitioning of retained energy

between body fat and protein, with less to adipose tissue and more to lean tissue. It is possible that the lower body fat content with the barley and wheat fed birds is related to lower rates of liver lipogenesis. Barley and wheat inclusion in diets affected the early growth of broilers, but the effect was largely alleviated in older birds. If barley and wheat are used alone or with corn in the growing stage (3-6 wk) only, the performance of broilers may be unaffected, but body fat content can be reduced compared with corn alone.

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CHAPTER VII

EFFECT OF DIETARY CEREAL ON BODY COMPOSITION, LIVER LIPOGENESIS AND BODY LIPID DEGRADATION IN BROILER CHICKENS

ABSTRACT

Five diets (corn, barley, wheat, barley + glucanase, and wheat + xylanase) with similar ME, protein, fat, starch and essential amino acid contents were fed to growing female broilers over the period of 3 to 6 wk to study the effect of dietary cereal, glucans and xylans on liver fatty acid synthesis. All the bird groups were controlled to have similar feed intake. In the growth study, all the birds had similar 6-wk body weight, but the corn-fed chickens contained significantly higher body fat than those fed barley, wheat and wheat-enzyme diet ($p<0.05$). Liver fatty acid synthase (FAS) and acetyl-CoA carboxylase (ACC) activities were higher for the chickens fed the corn diet than for those fed the barley and barley-enzyme diets, but were similar to those in chickens fed the wheat and wheat-enzyme diets. The incorporation of labeled water (^3H_2O) into liver fatty acids had a consistent pattern similar to that of FAS and ACC activities, but the difference among the diets was not significant ($p>0.05$). Plasma glycerol and free fatty acid concentrations were similar among the five bird groups. The lower feed intake in this experiment might have reduced the response of bird body composition, liver enzyme activities and liver fatty acid synthesis rate to the dietary differences. These results, together with the results in Chapter VI, indicate that dietary barley and wheat can reduce liver fatty acid synthesis, thus resulting in a lower body fat content in broiler chickens fed a barley or wheat based diet as compared with a corn diet. β-glucans and xylans in the diet did not seem to affect lipid metabolism.

Key words: cereal, fatty acid synthase, acetyl-CoA carboxylase, lipogenesis, body composition, broiler

INTRODUCTION

Chapter VI reported the effects of barley and wheat on body fat content of broiler chickens as compared with corn. Two trials were conducted in that study. In one trial, both the barley and the wheat diets significantly reduced bird body fat content at 6 wk of age as compared with the corn diet without affecting final body weight and other measures of performance. The barley diet had a greater effect on body fat content than the wheat diet. The barley and the wheat diets significantly changed the partition of stored energy, more to lean tissue and less to adipose tissue. The other trial confirmed the findings in the first trial and also produced a quantitative relationship between dietary barley or wheat inclusion level and chicken body fat content. Within limits, the higher the barley or wheat inclusion level in a diet, the lower the chicken body lipid content. The liver lipid content consistently changed with the body fat content, with a correlation coefficient of 0.74. Jensen et al. (1976) and Maurice and Jensen (1979) also observed that corn fed laying hens or Japanese quail had higher liver fat content and rate of fatty acid biosynthesis than wheat fed birds. Based on these results, it was assumed that the body fat content differences among the diets in our previous study are probably due to the differences in liver fatty acid synthesis. However, there have been no experiments reported which measured the effect of dietary barley or wheat on both body fat content and liver fatty acid synthesis at the same time with broilers or other animals.

Numerous studies have shown that some dietary fiber sources can lower blood total and low-density lipoprotein cholesterol, plasma triacylglycerol and triacylglycerol-rich lipoprotein concentrations (Cara et al., 1992; Kritchevsky & Story, 1993; Anderson et al., 1994; Lairon, 1996). The effect of dietary cereals on liver lipid metabolism may be related to their fiber type and its characteristics. Barley and corn contain a high proportion of water-soluble fiber (pectin and some hemicelluloses), whereas wheat contains higher proportions of insoluble fiber such as insoluble hemicelluloses, cellulose and lignin (MacGregor and Fincher, 1993; Betschart, 1988). The predominant non-starch polysaccharides of wheat endosperm are highly branched arabinoxylans, which have small amounts of phenolics associated with them. In contrast, barley endosperm cell wall is very rich in mixed-linkage β-glucans and relatively low in arabinoxylans. It was considered that some of these differences in physical and chemical properties of dietary carbohydrates might explain the fat-lowering effect of barley and wheat in our previous study.

However, reports regarding the effect of dietary pure fiber, such as β-glucans in barley and arabinoxylans in wheat, on liver fatty acid synthesis and body composition in broiler chickens are actually lacking. Therefore, the main objectives of this study were: (1) to further investigate the effects of barley and wheat on body composition compared with corn in broiler chickens using semi-purified diets; (2) to study the effects of barley and wheat on liver lipogenesis and plasma free fatty acid and glycerol levels compared with corn; and (3) to study the effect of β-glucans in barley and xylans in wheat on body composition, *in vivo* hepatic lipogenesis and plasma free fatty acid and glycerol concentrations.

MATERIALS AND METHODS

Experiment 7.1

Experimental design, dietary ingredients and composition

This is a single-factor completely randomized design with five dietary treatments. The main ingredients (corn, barley, wheat, soybean meal, isolated soybean protein, canola meal) were analyzed for crude protein, crude fat and moisture before they were used in diet formulation. Each diet was composed of the same percentage (62%) of either corn, barley or wheat (Table 7.1). Soybean meal, isolated soybean protein (Nurish Protein, St. Louis, MO), corn starch, corn oil (purchased in Safeway), tallow and synthetic methionine and lysine were used to balance dietary protein, energy, fat, methionine and lysine contents. The corn oil and tallow were used to maintain consistent saturation and content of dietary fatty acids among the diets. High pure β-glucanase (Betaglucanase Standard S126 47000 BGU/g) and xylanase (Endoxylanase Control C64 503000 EXU/g) were obtained from BASF Corporation, Wyandotte, MI, USA. The barley-enzyme diet was supplemented with BASF β-glucanase 5640 units/kg and the wheat-enzyme was supplemented with endoxylanase 5640 units/kg diet.

Animal experiment

Preliminary balance study: A preliminary balance study was conducted to measure apparent metabolizable energy (AME) to make sure the five diets were consistent in AME content. The total collection method was used. Forty day-old Hubbard female chicks were

Table 7.1. Diet composition and determined nutrient analyses (percentage unless stated) in Exp. 7.1

	Corn	Barley	Wheat	Barley+enzm[1]	Wheat+enzm[1]
Corn	62.0	...	...	...	...
Barley	...	62.0	...	62.0	...
Wheat	...	...	62.0	...	62.0
Soybean meal	21.8	6.0	13.6	7.3	16.4
Isolated soybean protein	5.0	10.0	5.0	9.7	3.4
Starch	6.0	16.3	12.4	15.7	11.1
Tallow	0.84	...	...	...	...
Corn oil	...	2.2	2.1	1.8	2.1
Limestone	1.14	1.48	1.88	1.48	1.88
Dicalcium phosphate	1.77	0.85	1.08	0.85	1.08
Iodized salt	0.62	0.35	0.87	0.35	0.87
Mineral premix[2]	0.25	0.25	0.25	0.25	0.25
Vitamin premix[2]	0.25	0.25	0.25	0.25	0.25
Rumensin premix[3]	0.10	0.10	0.10	0.10	0.10
DL-Methionine	0.06	0.02	0.11	0.02	0.12
L-Lysine.HCl	0.11	0.10	0.23	0.10	0.27
L-Threonine	0.10	0.11	0.15	0.11	0.21
Total	100	100	100	100	100
Determined analyses					
Dry matter	88	90	88	90	88
AME, kcal/kg	3129	3097	3068	3064	3062
Crude protein	19.3	19.1	19.8	19.1	19.9
Crude fat	4.3	4.3	4.1	4.1	4.1
Starch	44.2	43.3	41.1	40.7	43.0
Lysine[4]	1.00	1.00	1.00	1.00	1.00
Methionine[4]	0.39	0.39	0.39	0.39	0.39
Met + Cys[4]	0.67	0.69	0.68	0.70	0.68
Threonine[4]	0.74	0.74	0.74	0.74	0.74
Tryptophan[4]	0.21	0.24	0.22	0.25	0.22
Calcium	1.00	0.85	1.20	0.86	1.10
Available phosphorus[4]	0.47	0.35	0.35	0.35	0.35
NDF[5]	6.6	15.2	10.5	12.8	9.1
ADF[5]	4.4	6.8	4.6	6.1	4.3

[1] The barley-enzyme and the wheat-enzyme diets were supplemented with BASF β-glucanase and endoxylanase 5640 units/kg diet, respectively.

[2] The same as in Table 6.2 in Chapter VI.

[3] Contaning monensin sodium 200 g/kg, Elaanco Division, Eli Lilly Canada, Inc., Guelph, Ontario, Canada

[4]Calculated; Met + Cys: methionine + cystine

[5]NDF: neutral detergent fiber; ADF: acid detergent fiber.

obtained from a commercial hatchery and allocated to 10 cages, 4 birds/cage, in an animal room. All the birds were fed a commercial starter diet up to 3 wk of age. At 3 wk of age, the birds in the 10 cages were fed the five test mash diets, with two cages per diet. After a wk of adaptation to the diets, feed intake and excreta were measured for 3 d. The diet and excreta samples were

analyzed for dry matter and gross energy, and the diet samples were analyzed for crude protein, crude fat, and starch contents.

Growth study: Ninety day-old Hubbard female chicks with similar body weight were obtained from a commercial hatchery and were allocated randomly to 20 cages in the University Avian Center. All the chicks were fed the same commercial starter diet during the first 3 wk. At 3 wk of age, 80 birds selected were reallocated using random block method to the 20 cages based on their weights. The five diets were assigned to the 20 cages with four replicate cages and a total of 16 birds for each diet. The lighting program was 18 h light and 6 h dark. The light intensity was 20 lx during first 3 d and 5 lx thereafter. The room temperature was maintained the same as in previous experiments.

The same amount of feed was weighed daily for each cage with four birds. The birds fed the corn and wheat diets did not eat to their appetite, while the birds fed the barley diets consumed as much as they could maximally daily since the barley mash diets had poor palatability. The feed amount was gradually increased with age. At 6 wk of age, birds were weighed individually after an 8 h fasting. Subsequently, the birds were killed by cervical dislocation and the liver was excised, weighed and frozen immediately in liquid nitrogen. Then the liver was moved to a freezer of -70 C for later liver enzyme activity analyses. The remaining carcasses were frozen at –20 C for body composition determination.

Isotope Study: Another 70 day-old Hubbard female broiler chicks were allocated to 10 cages in an animal laboratory, with 5-6 birds per cage at the beginning. The birds were fed a commercial starter diet for the first 3 wk. At 3 wk of age, 50 birds of relatively consistent body weight were relocated to 10 cages using a random block method, with five birds per cage. The birds were fed the five test diets during 3-6 wk, with two cages and 10 birds per diet. Feed intake was controlled the same way as in the growth study. A balance study was conducted within the fifth week, similar to the procedure in the preliminary balance study.

At 37 days of age, birds were fasted for 15 hours and weighed individually. A 3 ml blood sample was collected into a heparinized vacuum tube from a wing vein. The blood samples were put on ice after collection. Plasma samples were prepared by centrifuging at 3000 × *g* for 10 min and stored at –20 C for later non-esterified fatty acid and glycerol analyses. Two days later, a catheter was inserted to another wing vein and an adapter was screwed into the outside the end of catheter. After bandaging, the bird was put back to the original cage for 1 d adaptation to the

catheter implantation. On the second day, 100 μCi tritiated water/ml saline (0.9%) was injected through the catheter adapter at 100 μCi/kg body weight. Thirty min after the ^3H_2O injection, the birds were killed by cervical dislocation. The liver was removed immediately, weighed, and frozen in liquid nitrogen, and thereafter moved to a -70 C freezer for later ^3H-fatty acid determination. The bird carcasses containing ^3H_2O were disposed of based on the University Radionuclide Safety Guide.

Laboratory and statistical anslyses

Bird body composition (water, ash, and lipids), dry matter, gross energy and nitrogen, in dietary and excreta samples, calcium, phosphorus, neutral detergent fiber and acid detergent fiber in dietary samples were analyzed as in previous trials. Dietary and body fatty acid profile was analyzed using the method of Folch et al. (1957) to extract the lipids followed by saponification using 0.5 N methanol-KOH at 50 C for 1 h. The non-saponifiables were removed with 2.5 ml petroleum ether. Two drops of 0.4 N HCl and 5 ml BF_3 were added and boiled in boiling water for 15 min. The fatty acid methyl esters were extracted with 2.5 ml hexane. One ml top hexane layer was transferred into an eppendorf tube whose bottom barely covered with a drying agent (Na_2SO_4: $NaHCO_3$, 4:1). One ul of the hexane extract was injected to the Shimadzu GC-17A (Shimadzu Scitific Inc., Ltd, Japan) for fatty acid analysis. Plasma glycerol was determined using a Glycerol UV Test kit (Boehringer Mannheim, Italy) and non-esterified fatty acid concentrations were determined using a Wako NEFA C test kit (Wako Chemicals USA, Inc.). HCl-fat in both diet and excreta samples were analyzed using the method of Wiseman et al. (1992). Dietary and excreta starch was hydrolyzed to glucose using the procedure of Holm et al. (1986), then quantified by using a glucose assay kit (Sigma Chemicals). Excreta uric acid content was determined based on Marquardt (1983). Digestibility of dietary protein was estimated as [nitrogen intake - (total excreta nitrogen - uric acid nitrogen)]/nitrogen intake × 100, as uric acid nitrogen accounts for approximately 88% of the urinary nitrogen (Krogdahl and Dalsgard, 1981) in poultry.

The rate of ^3H incorporation from ^3H_2O into fatty acids in the liver was determined based on Griffin et al. (1992), Saadoun and Leclercq (1983) and Donaldson (1985). Liver lipids were extracted based on Folch et al. (1957). The lipid extract in chloroform was evaporated under a nitrogen stream. The lipid weight was recorded. The lipids were saponified with 10% KOH in

70% alcohol at 60-70 C. Unsaponifiable lipids were removed with hexane. The aqueous phase was acidified with HCl and the fatty acids were removed by three successive extractions with 4-ml hexane. The combined hexane extracts were back-washed with water three times and evaporated under a nitrogen stream. The fatty acids were weighed and dissolved in 6 ml of toluene-based scintillation cocktail (3.92 g 2,5-diphenyloxazole, 80 mg *p-bis*-(*o*-methylstyryl)-benzene, 230 ml ethyl alcohol and toluene to 1 liter) before counting in an LS 6500 Multipurpose Scintillation Counter, Beckman Instruments, Inc., Fullerton, CA. Results were expressed as pCi ^3H_2O incorporated into liver fatty acids in 30 min per μCi ^3H_2O injected into the body.

Enzyme activity assay: the activity of acetyl-coenzyme A carboxylase (EC. 6.4.1.2) in the liver was assayed based on Lopaschuk et al (1994) and Thampy and Wakil (1985). Samples of 1-g frozen liver were homogenized on ice with 8 ml of extraction buffer containing 50 mM tris (hydroxymethyl) aminomethane (Tris)-HCl (pH 7.5 at 4 C), 50 mM NaF, 0.25 M mannitol, 1 mM ethylenediamine-tetraacetic acid (EDTA), 1 mM ethylene glycol-bis (b-aminoethylether)-N,N,N',N'-tetraacetic acid (EGTA), 1 mM dithiothreitol (DTT), 5 mM sodium pyrophosphate, 1 mM phenylmethanesulphonyl fluoride (PMSF), 1 mM benzamidin and 4 ug/ml each of soybean trypsin inhibitor, aprotinin, leupeptin, and pepstatin A. Homogenates were centrifuged at 14,000 × *g* (Sorrall RC-5B Refrigerated Superspeed Centrifuge, Du Pont Instrument, Newtown, CT) for 20 min at 4 C. PEG 8000 was added to the supernatant so that it contained 2% PEG 8000, stirred for 10 min at 4 C and then centrifuged at 10,000 × *g* for 10 min. Three ml of the supernatant and 1 ml of 40% PEG 8000 was mixed on ice and centrifuged at 10,000 × *g* for 10 min to precipitate acetyl-CoA carboxylase protein. The pellet was re-suspended in 3 ml of re-suspension solution containing 100 mM Tris-HCl (pH 7.5 at 4 C), 1 mM EDTA, 1 mM EGTA, 1 mM DTT, 50 mM NaF, 5 mM Na pyrophosphate, 10% glycerol, 0.02% sodium azide, 4 ug/ml soybean trypsin inhibitor and 1 mM benzamidine. The protein content of the enzyme solution was measured based on Bradford (1976). The assay solution was composed of 60.6 mM tris-acetate, 2.12 mM ATP, 1.32 uM b-mercaptoethanol, 5.0 mM Mg acetate, 10 mM K citrate, 1.06 mM acetyl-CoA, 18.18 mM $NaHCO_3$, 0.33 uCi/umol $NaH^{14}CO_3$ and 1 mg/ml fatty acid free bovine serum albumin (pH 7.5), in which ATP and acetyl-CoA solutions were made freshly. The reaction was started by adding 20 ul of enzyme solution (pre-incubated for 5 min) in a final assay mixture of 330 ul. After 5 min of incubation at 37 C, the reaction was stopped by addition of 50 ul of 10% perchloric acid. Reaction tubes (Micro-centrifuge tubes) were immediately placed in a

dessicator under vacuum to remove un-reacted label as $^{14}CO_2$. The tubes were centrifuged at 2900 × *g* for 20 min with a desktop centrifuge (Bioguge 13, Heraeus Instruments, Germany). 320 ul of the supernatant was transferred into a 7-ml plastic scintillation vial and evaporated to dryness at 80 C. The residue was dissolved in 100 ul of H_2O and mixed with 6 ml of Ready Safe™ liquid scintillation cocktail (Beckman) for determination of radioactivity. Acetyl CoA carboxylase activity was expressed as nmol of ^{14}C-bicarbonate incorporated into malonyl-CoA.min^{-1} mg protein^{-1}.

Fatty acid synthase activity (FAS) was assayed based on Bannister et al. (1983) and Arslanian and Wakil (1975). Samples of 1.5 g liver were homogenized for 45 sec in 10 ml of homogenate buffer (pH 7.4) containing 0.05 M K phosphate buffer, 1 mM dithiothreitol (DTT) and 1 mM EDTA. The homogenate was centrifuged at 15,000 × *g* for 10 min at 4 C and 5 ml of the supernatant was transferred to a 13-ml Beckman UltraClear tube. Another 6 ml of homogenate was added to the tube before centrifuging at 105,000 × *g* for 1 h at 4 C using a Beckman ultra centrifuge (Beckman Instruments Inc., Irvine, CA). The supernatant was measured for protein content based on Bradford (1976) and was used for the enzyme assay. Enzyme solution (0.2 ml) was pre-incubated with 0.7 ml assay solution in a plastic cuvette at 37 C for 5 min prior to reading the absorbance (A1) at 340 nm. The assay solution contained four portions of 188 mM K phosphate buffer (pH 6.5), four portions of 12.5 mM DTT, one portion of 0.25 mM acetyl-CoA (made freshly) and one portion of 1.5 mM NADPH (made freshly). Malonyl-CoA solution was added to start the reaction. After incubation at 37 C for exactly 10 min, the absorbance (A2) was read immediately at 340 nm. Reference (blank) was treated the same except that the enzyme solution was replaced by homogenate buffer. Specific activity of FAS was expressed as nmol NADPH oxidized min^{-1} mg protein^{-1}.

$$\text{FAS (nmol/min/mg protein)} = (V \times \Delta A) / (\varepsilon \times d \times v \times cp) = 852090\ \Delta A / cp$$

Where V - final volume (1.06 ml) in the assay; v - sample volume (0.2 ml), d - light path (1 cm), ε - extinction coefficient of NADPH (6.22 mM $^{-1}$ cm^{-1} at 340 nm), cp - protein concentration in the sample enzyme solution (ug/ml), ΔA - $[(A_{sample1} - A_{blank1}) - (A_{sample2} - A_{blank2})]$/10 min.

All parometers were analyzed statistically using analysis of variance for a completely randomized design. Any significant differences between the diets were further compared using Duncan's multiple range test (SAS Institute Inc., 1996).

RESULTS

Dietary Nutrient Contents and Digestibility

All five diets contained similar metabolizable energy, crude protein, crude fat, starch, lysine, methionine, threonine, cystine, and tryptophan, calcium, and phosphorus as shown in Table 7.1. Fatty acids profile was similar among the five diets except that the corn diet was lower in C18:2 and higher in 16:1, C18:0 and C18:1 than other diets which were similar (Table

Table 7.2 Dietary and carcass selected fatty acids (relative percentage of total fatty acids, Exp. 7.1)

Fatty acid	C14:0	C16:0	C16:1	C18:0	C18:1	C18:2	C18:3	C20:0	C22:0	P/S ratio[3]
					Diet[1]					
Corn diet		21.8	3.4	8.7	38.8	25.3	1.3	0.4	0.3	0.85
Barley diet		18.7	2.3	4.9	31.8	39.0	2.4	0.3	0.6	1.69
Wheat diet		21.4	0.7	5.3	32.1	37.5	2.0	0.5	0.5	1.43
Barley-enzyme		21.8	1.1	5.4	34.3	34.1	2.1	0.5	0.6	1.28
Wheat-enzyme		22.0	1.2	6.8	32.2	34.6	2.1	0.6	0.4	1.23
					Carcass					
Corn diet	0.6	24.5	7.3[a]	7.4	44.7	13.9[b]	0.8	0.2	0.7	
Barley diet	0.6	24.3	6.0[b]	7.9	42.6	16.9[a]	1.0	0.1	0.7	
Wheat diet	0.6	24.7	6.4[ab]	7.9	42.6	16.3[a]	0.9	0.1	0.6	
Barley-enzyme	0.6	24.6	5.9[b]	8.0	42.6	16.3[a]	1.1	0.2	0.7	
Wheat-enzyme	0.6	24.7	6.7[ab]	7.8	42.5	16.2[a]	0.9	0.2	0.5	
SEM[2]	0.0	0.5	0.2	0.2	0.8	0.5	0.1	0.0	0.0	
Significance			**			*				
N[2]	3	3	3	3	3	3	3	3	3	

[ab]. Means with different supercripts are significantly different, * $p<0.05$; ** $p<0.01$.

[1] Stastistical analysis of dietary fatty acid profile was not available.

[2] SEM: standard error of mean; N: number of observations.

[3] P/S ratio: polyunsaturated fatty acids/saturated fatty acids.

Table 7.3 Digestibility of the dietary starch, fat and protein and nitrogen retention rate (%)

Diet	Dry matter digestibility	Starch digestibility	Fat digestibility	Protein digestibility	Nitrogen retention
Corn	76.7[a]	100.0	85.7[b]	94.3[a]	59.0[a]
Barley	66.5[e]	100.0	81.9[c]	80.2[c]	50.8[c]
Wheat	73.3[c]	99.9	86.3[b]	88.3[b]	55.6[ab]
Barley-enzyme	68.8[d]	100.0	88.1[ab]	82.3[c]	52.0[bc]
Wheat-enzyme	75.1[ab]	100.0	91.1[a]	87.5[b]	58.6[a]
SEM	0.3	0.1	0.9	1.1	1.2
Significance	**	n.s.	*	*	*
N	2	2	2	2	2

[abcd] Means with different superscripts are significantly different, * $p<0.05$; ** $p<0.01$.
n.s. not significantly different.

7.2). The barley and the barley-enzyme diets contained higher fiber as neutral detergent fiber and acid detergent fiber than the other three diets (Table 7.1).

Among the five diets, the two barley diets had the lowest dry matter digestibility followed by the two wheat diets, while the corn diet had the highest dry matter digestibility (Table 7.3). Supplementation of the barley and the wheat diet with β-glucanase and xylanase, respectively, significantly improved their dry matter and fat digestibility ($p<0.05$, Table 7.3) as compared with their counterparts without the enzymes. Starch digestibility was 100% for all five diets. The protein digestibility of the corn diet was the highest and that of the barley and barley-enzyme diets was the lowest, while the protein digestibility of the two wheat diets was intermediate ($p<0.05$). Supplementation of enzymes did not significantly ($p>0.05$) improve protein digestibility of the barley-enzyme and the wheat-enzyme diet. The nitrogen retention rate was lower for the barley diets as compared with the other three diets which did not differ ($p>0.05$).

Performance

The average final body weights of the chickens in the growth study were not significantly different among the five diet groups ($p>0.05$, Table 7.4). Basically, all the chicken groups had similar feed intake. Chickens fed the diets supplemented with enzymes had slightly lower feed: gain ratio than their counterparts without enzymes (Table 7.4). AME: gain ratios for the

Table 7.4 Effect of diets on growth performance of broiler chickens in the growth study (Exp. 7.1)

Diet	Body wt[1] g/bird	Body wt gain[2] g/bird	Feed intake g/d/bird	Feed: gain g/g	AME/gain cal/g
Corn	1482	864	83.3	1.70[a]	5309[a]
Barley	1466	815	82.2	1.71[a]	5305[a]
Wheat	1507	887	83.3	1.65[ab]	5069[ab]
Barley-enzyme	1472	823	82.2	1.69[a]	5192[ab]
Wheat-enzyme	1537	916	83.3	1.60[b]	4904[a]
SEM	43	35	0.8	0.03	119
Significance	n.s.	n.s.	n.s.	*	*
N	16	16	4	4	4

n.s: not significant ($p>0.05$);
[ab] Means with different superscripts are significantly different, * $p<0.05$.
[1] Body weight at 6 wk of age.
[2] Body weight gain during 3-6 wk.

enzyme-supplemented diets also tended to be lower than their counterparts without enzymes, but the difference was not significantly ($p>0.05$). The birds fed the corn, barley and barley-enzyme diets had a significantly higher feed: gain ratio than those fed the wheat-enzyme diet ($p<0.05$).

Body Composition

The body fat content of the corn-fed chickens was significantly higher ($p<0.05$) than that of the birds fed the barley, wheat, and wheat-enzyme diets at 6 wk of age (Table 7.5). The body water, protein and ash contents were similar among the five bird groups. The carcass energy content of the corn-fed chickens was significantly higher than that of the chickens fed the wheat-enzyme diet ($p<0.05$, Table 7.5). The carcass fatty acid profile was similar among the five diets except that the birds fed the corn diet contained lower C18:2 ($p<0.05$) than those fed the four test diets and higher C16:1 ($p<0.01$) than the birds fed the two barley diets (Table 7.2).

Table 7.5 Effect of diets on body composition in the growth study (Exp. 7.1)

Diet	Water, %	Fat, %	Protein, %	Ash, %	Carcass energy, cal/g
Corn	67.2	11.1^{a}	19.0	2.68	2086^{a}
Barley	67.8	10.5^{b}	19.0	2.75	2027^{ab}
Wheat	67.9	10.5^{b}	18.9	2.71	2024^{ab}
Barley-enzyme	67.3	10.7^{ab}	19.3	2.75	2062^{ab}
Wheat-enzyme	68.1	10.4^{b}	18.8	2.71	2009^{b}
SEM	0.3	0.2	0.2	0.04	22
Significance	n.s.	*	n.s.	n.s.	*
N	16	16	16	16	16

^{ab} Means with different superscripts within a column are significantly different, *: $p<0.05$.
n.s.: not significantly different, $p>0.05$.

Lipogenic Enzyme Activities

Wheat fed birds tend to be higher in liver weight than the other bird groups, but only the birds fed wheat-enzyme diet had significantly higher liver weight than that of the birds fed the barley diet ($p<0.05$, Table 7.6). Birds fed the barley and barley-enzyme diets showed lower FAS and ACC activities ($p<0.05$ in some comparisons, Table 7.6) than those fed the corn, wheat or wheat-enzyme diet. ACC activities in wheat fed birds were intermediate.

Body Fat Degradation and Liver Fatty Acid Synthesis

Plasma glycerol and non-esterified fatty acid concentration of chickens in the starved state were determined to reflect the rate of lipolysis (lipid degradation). There were no significant differences in plasma glycerol and non-esterified fatty acid concentrations among the five diet groups ($p>0.05$, Table 7.7). Plasma glycerol and non-esterified fatty acid concentrations of the five diet groups were highly correlated ($r=0.85$, $p<0.05$).

Table 7.6 Liver fatty acid synthase and acetyl-CoA carboxylase activities in the liver (Growth study)

Diet	Liver weight, g	FAS-p[1]	FAS-liv[2]	ACC-p[3]	ACC-liv[4]
Corn	35.6^{ab}	543^{a}	19.4^{a}	1.55^{a}	913^{a}
Barley	35.1^{b}	470^{c}	16.6^{b}	1.28^{b}	711^{c}
Wheat	39.0^{ab}	513^{abc}	19.2^{a}	1.39^{ab}	860^{ab}
Barley-enzyme	35.8^{ab}	482^{bc}	16.9^{b}	1.21^{b}	755^{bc}
Wheat-enzyme	40.9^{a}	517^{ab}	19.3^{a}	1.41^{ab}	884^{a}
SEM	1.8	15	0.6	0.08	41
Significance	*	*	*	*	*
N	16	16	16	16	16

[1] Liver fatty acid synthase activity: nmol NADPH. min^{-1}. mg $protein^{-1}$

[2] Liver fatty acid synthase activity: mmol NADPH. min^{-1}. $liver^{-1}$

[3] Acetyl-CoA carboxylase activity: nmol ^{14}C-$NaHCO_3$ incorporated into malonyl-CoA. min^{-1}. mg $protein^{-1}$.

[4] Acetyl-CoA carboxylase activity: nmol ^{14}C-$NaHCO_3$ incorporated into malonyl-CoA. min^{-1}. $liver^{-1}$.

[abc] Under each column, means followed by different letter superscripts are significantly different ($p<0.05$).

The chickens fed the wheat and the wheat-enzyme diets tended to contain higher liver lipid and fatty acid contents than those fed the barley and the barley-enzyme diets but the difference was not significant ($p>0.05$, Table 7.7). There was no significant differences ($p>0.05$) in the rates of liver lipogenesis expressed as pCi ^3H_2O incorporated into liver fatty acids over 30 min per μCi ^3H_2O injected into the wing vein among the five bird groups (Table 7.7). The pattern of liver fatty acid synthesis rate between diets is similar to that of FAS and ACC activities. The correlation coefficients between FAS activity (per mg protein) and fatty acid synthesis rate and between ACC activity (per mg protein) and fatty acid synthesis rate were 0.89 ($p<0.05$) and 0.95 ($p<0.01$), respectively

DISCUSSION

Nutrient Digestibility

Non-starch polysaccharides in barley and wheat diets produce viscosity in the lower portion of the small intestine and impair diffusion of pancreatic enzymes and their substrates (Annison and Choct, 1991), thus reducing the dry matter and fat digestibility of the diets containing barley and wheat. In addition, soluble fiber including β-glucans and arabinoxylans is able to combine bile acid in the intestinal tract, thus reducing emulcification of dietary fat and fat digestibility (Lairon, 1996). The glucanase added to the barley-enzyme diet and the xylanase to

Table 7.7 Fatty acid synthesis and plasma glycerol and non-esterified fatty acids (Exp. 7.1)

Diet	Glycerol, mg/L	NEFA[1], mmol/L	Liver lipids, %[2]	Liver FA, %[2]	Lipogenesis[3]
Corn	10.6	0.65	7.4	3.6	39.7
Barley	9.7	0.54	7.1	2.9	36.6
Wheat	10.3	0.56	8.2	3.8	38.1
Barley-Enzyme	10.1	0.53	6.7	3.3	35.9
Wheat-Enzyme	10.8	0.63	8.4	3.7	37.2
SEM	0.8	0.04	0.9	0.5	4.3
Significance	n.s	n.s.	n.s.	n.s.	n.s.
N	15	15	8	8	8

[1] NEFA: non-esterified fatty acids.

[2] On fresh weight basis.

[3] Liver fatty acid synthesis rate was expressed as *p*Ci ^3H_2O incorporated into liver fatty acids over 30 min per μCi ^3H_2O injected into the wing vein.

n.s.: not significantly different, $p>0.05$.

the wheat-enzyme diet, to a great extent, broke the non-starch polysacharide network type structure and significantly improved the dry matter and fat digestibility ($p<0.01$, Table 7.3).

The dietary non-starch polysaccharides including β-glucans and xylans did not affect starch digestibility of 3-6 wk old broiler chickens. McNab (1992) stated that in adult birds starch is also completely digested. The protein digestibility of the corn diet was the highest and that of the barley and the barley-enzyme diets was the lowest, while the protein digestibility of the two wheat diets were intermediate. Supplementation of enzymes did not significantly ($p>0.05$) improve protein digestibility, suggesting that dietary fiber other than β-glucans and xylans in barley and wheat might be responsible for the differences in protein digestibility among the diets. The nitrogen retention rate had a similar pattern to the protein digestibility, indicating that the difference in nitrogen retention was due mainly to the protein digestibility difference.

Growth Performance

The average final body weight of chickens in the growth study was not significantly different among the five diet groups ($p>0.05$, Table 2). Although non-significantly ($p>0.05$) different, body weight of birds fed the two wheat diets tended to be higher than that of birds fed the corn, barley and barley-enzyme diets. This pattern was quite similar to the result observed in one of our previous experiments (Chapter VI). Pearson et al. (1978) also reported that the final body weight of laying hens followed the order wheat > maize > barley when the diets based on maize, wheat and barley respectively, were fed to pullets for 13 wk from point of lay.

AME: gain ratio indicates dietary AME requirement for a unit of weight gain and reflects the utilization of dietary energy for body weight gain. Generally, the corn-fed birds had a higher AME: gain ratio than other bird groups, due probably to the higher body fat content in the corn-fed chickens as discussed in Chapter VI. The birds fed the barley and the barley-enzyme diets had higher feed: gain and AME: gain ratios than the wheat fed birds, reflecting the fact that the barley diets had more serious growth-depressing effect than the wheat diets as mentioned earlier (Chapter VI). Those fed the diets supplemented with enzymes had a lower feed: gain and AME: gain ratio than their counterparts without enzymes (Table 7.4), indicating that the supplemental enzymes removed, at least partially, the effect of the anti-nutrients.

Body Composition

Consistent with the results in our previous experiments (Chapter VI), the barley, wheat and wheat-enzyme diets significantly reduced body fat content without affecting final body weight as compared with the corn diet ($p<0.05$). The body lipid reduction in the current experiment was not so pronounced as in our previous two experiments (Chapter VI), due probably to the lower feed intake in all the bird groups in the current study. The chickens fed the corn diet deposited more fat thus more energy than birds in the other groups, indicating that the corn-fed birds utilized the dietary energy more efficiently for body fat and energy deposition. This is consistent with the findings in our previous experiments (Chapter VI).

Chickens fed the wheat and wheat-enzyme diets had similar body fat content to those fed the barley and barley-enzyme diets. This result does not agree with that observed in Chapter VI

in which the chickens fed the barley diets contained lower body fat than those fed the wheat diets. The main reason for this difference is probably that the feed intake of birds in the current study was just 60% of that in a previous experiment (Chapter VI) with the same sex of birds. The lower feed intake for all the bird groups minimized the achievement of body fat growth potential, thus minimizing the difference in body lipid content between the diet groups. Even though, the birds fed the corn diet still showed higher body lipid than those fed the other diets.

Lower C18:2 and higher C16:1 in the corn diet resulted in lower C18:2 ($p<0.05$) and higher C16: 1 ($p<0.01$) contents in the corn fed birds than in the birds fed the other diets. This result was consistent with that of Hulan et al. (1988). Higher saturated fatty acid C18: 0 in the corn diet did not increase the percentage of C18:0 in the birds fed the corn diet as compared with the birds fed the other diets, probably due to the desaturation of saturated fatty acids after they are absorbed (NRC, 1994). In mammals, lower dietary PUFA was reported to increase body fat content (Shimomura et al., 1990). However, in the current study, the lower C18:2 and C18:3 and higher C18:1, 18:0 and C16:1 (i.e., lower PUFA/SFA ratio) in the corn diet is probably not responsible for the higher body fat and FAS and ACC activity in birds fed the corn diet compared with the barley diets. The PUFA/SFA ratio was similar between the barley and wheat diets in the current experiment, but FAS and ACC activities were significantly different between birds fed the two diets ($p<0.05$, Table 7.6). Also, in Exp. 6.1 in Chapter VI, the barley and wheat diets supplemented with higher tallow-soya oil (1:1) blend and presumably containing higher PUFA/SFA ratio, resulted in lower body fat content as compared wth the corn diet ($p<0.01$). Therefore, PUFA/SFA ratio difference among diets may not be responsible for the bird body fat and FAS and ACC activity differences in broilers in Chapter VI and VII. No significant differences ($p>0.05$) were observed in the body fat accumulation of broilers fed diets rich in PUFA than rich in SFA in the literature (Hulan et al., 1988; Olomu and Baracos et al., 1991).

The body protein and ash contents were similar among the five diet groups. Supplementation of β-glucananse or xylanase to the barley-enzyme or the wheat-enzyme diets, respectively, did not affect the fat-reducing effect of the barley and wheat.

Hepatic Lipogenic Enzyme Activities and Fatty Acid Synthesis

The activities of liver fatty acid synthase (FAS) and acetyl-CoA carboxylase (ACC) of birds from the growth study were determined to indicate liver fatty acid synthesis rate. The two

enzymes are key enzymes in the process of fatty acid synthesis. As listed in Table 7.6, the chickens fed the corn diet had higher FAS and ACC activities expressed as per mg protein or per liver than those fed the barley or barley-enzyme diet ($p<0.05$). This is consistent with the body fat content result listed in Table 7.5, suggesting that the body fat content difference resulted from the difference of liver fatty acid synthesis.

Maurice and Jensen (1979) observed that the laying hens fed a corn based diet had significantly higher hepatic FAS activity as compared with the birds fed a wheat based diet. In the current study, FAS activity was similar between the birds fed the corn and the wheat diet. The inconsistence is possibly that the low feed intake in the current study reduced the difference of FAS activity between the diets.

In one of our previous experiments (Exp. 6.1, Chapter VI), it was found that corn-fed chickens had higher liver lipid content than those fed a barley or a wheat diet. However, in the current study, the liver lipid content was similar among the diet groups, although chickens fed the two wheat diets tended to have higher liver weight and liver lipid contents than the corn- or barley-fed chickens. This difference is not clear. Qureshi et al. (1980) reported similar liver weights between chicks fed a corn diet and those fed a wheat diet at 3 wk of age.

The incorporation of labeled water (^3H_2O) into liver fatty acids among the five diet groups was not significantly different ($p>0.05$). Maurice and Jensen (1977) reported that incorporation of [1-^{14}C]-acetate into liver lipid *in vivo* was significantly higher in birds fed a corn diet than those fed a wheat diet. *In vitro* incorporation of ^{14}C into liver lipid was significantly elevated in the liver samples from birds fed a corn diet as compared with the birds fed a wheat diet. Maurice and Jensen (1979) also reported that Japanese quail fed a wheat diet for 6 wk significantly reduced the rate of liver fatty acid synthesis measured with [1-^{14}C] acetate as compared with those fed a corn diet. The reason for this difference is probably the large variability of measured lipogenesis within each group in the current study. Although not significantly different, the pattern of fatty acid synthesis rate measured using ^3H_2O among diets was similar to that of FAS and ACC activieies. Also the fatty acid synthesis rate was highly correlated with FAS or ACC activities ($r=0.89$ and 0.95, respectively). Again, the relatively low feed intake accounts for the small differences which are consistent with the FAS and ACC activity data.

Supplementation with dietary enzymes did not affect FAS and ACC activities and the incorporation rate of ^3H_2O into liver fatty acids in the current study. These results indicate that

dietary β-glucans and xylans did not affect liver lipid metabolism and body fat deposition. To the author's knowledge, there have not been similar reports regarding the effect of dietary β-glucans and xylans on liver FAS and ACC activities reported in the literature.

Plasma Glycerol and Non-Esterified Fatty Acids

Plasma glycerol and non-esterified fatty acids (free fatty acids) of chickens in the starved state were determined to reflect the rate of body lipid degradation. There were no significant differences ($p>0.05$) in plasma glycerol and non-esterified fatty acid concentrations among the five diet groups (Table 7.7), indicating that the dietary treatments did not affect body lipolysis in the current study. Pearson et al. (1978) also did not observe any difference in plasma non-esterified fatty acid among birds fed corn, wheat or barley based diets. Plasma glycerol and non-esterified fatty acid concentrations of the five groups were highly correlated ($r=0.85$), indicating that glycerol and non-esterified fatty acid concentrations in the plasma were consistent. The addition of enzymes to the barley-enzyme and wheat-enzyme diets did not affect plasma glycerol and non-esterified fatty acid concentrations, indicating that β-glucans and xylans do not affect body lipid degradation.

CONCLUSION

In the present study, the chickens fed the barley, the wheat, and the wheat-enzyme diet had significantly lower body fat content than those fed the corn diet, but the body weights were similar among the five dietary groups. The activities of key enzymes (FAS and ACC) in the liver fatty acid synthesis process were higher for the chickens fed the corn diet than for those fed the barley and barley-enzyme diets, but were similar to those in chickens fed the two wheat diets. Incorporation of labeled water into liver fatty acids had a consistent pattern similar to that of the key enzyme activities. These observations, together with our previous results in Chapter VI, lead to this conclusion: the barley or wheat in a diet reduced liver fatty acid synthesis, thus resulting in a lower body fat content in broiler chickens fed the barley or the wheat based diet. Dietary β-glucans and xylans do not seem to affect lipid metabolism in broiler chickens.

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CHAPTER VIII

INFLUENCE OF LIGHTING PROGRAMS ON PERFORMANCE AND BODY FAT CONTENT OF BROILER CHICKENS

ABSTRACT

In two trials, effects of lighting programs on performance and body composition were evaluated. Exp. 8.1 was a 2 diet × 4 lighting program factorial experiment (the same as Exp. 4.2 in Chapter IV). The four lighting programs were control (CON1, 18L: 6D), increasing (INC1), intermittent (INT) and increasing-intermittent (INC-INT) lighting programs. A total of 2400 male Peterson × Arbor Acre day-old chicks was raised to 42 d of age in 16 pens. Among the three test lighting programs (INC1, INT and INC-INT), only INC1 significantly reduced the feed intake and body weight gain during 0-3 wk ($p>0.05$), but INC1 did not result in compensatory growth or change of body composition during 3-6 wk as compared with CON1 birds.

Exp. 8.2 was a factorial experiment of 3 diet × 4 lighting program × sex design (the same as Exp. 6.1 in Chapter VI). A total of 2800 Peterson × Arbor Acre chicks was raised to 42 d of age. The four lighting programs were control (CON2, 16L: 8D), and three increasing lighting schedules (INC2, INC3, and INC4). INC2, INC3 and INC4 reduced feed intake and body weight gain during 0-3 wk ($p<0.05$) and the birds kept under INC2, INC3 and INC4 obtained (nearly) complete compensatory growth by 6 wk as compared with those under CON2. The feed: gain ratio was significantly lower for INC2, INC3 and INC4 during 0-3 wk, and for INC4 during 3-6 wk than for CON2. INC2, INC3 and INC4 resulted in significantly higher body water and protein ($p<0.05$) and lower lipids (but $p<0.11$) at 6 wk as compared with CON2. The SDS mortality for INC2, INC3 and INC4 birds was lower than for CON2 birds, but only significantly lower for INC4 birds. The results suggest that the three test lighting programs (especially INC4) are fairly successful in lowering body lipid accretion and incidence of metabolic diseases such as SDS, and maintaining final body weight.

Key words: lighting, body weight, body composition, sudden death syndrome, broilers

INTRODUCTION

Lighting and Performance, Metabolic Diseases and Behavior

Traditionally, a low-intensity, continuous or near continuous light has been provided to maximize feed intake, and thus growth rate (Buyse et al., 1996b). Since modern broiler chickens, capable of rapid growth, exhibit a high incidence of metabolic and skeletal diseases (Charles et al., 1992) and a high body fat content (Whitehead, 1988), researchers have been trying to modify the pattern of growth, health and body composition of broilers through altering lighting pattern.

It was reported that the incidence of metabolic and skeletal diseases (e.g., sudden death syndrome and ascites) could be reduced through reducing the photoperiod (feeding time) in various lighting programs for birds at an early age without affecting their final body weight (Classen and Riddell, 1989; and Blair et al., 1993). The increasing and intermittent lighting programs are believed to reduce feed intake, slow down early growth and increase bird activity, thus reducing the incidence of metabolic diseases associated with a rapid growth but without affecting skeletal development. The skeleton may then be more capable of supporting rapid increase in live weight when the photoperiod is subsequently increased (Classen and Riddell, 1989; Gorden, 1994). Upon restoration of long photoperiod, the birds obtained accelerated growth and achieved a similar final body weight to the control birds in many studies (Classen and Riddell, 1989; Blair et al., 1993).

Birds given an intermittent light program mainly limit feeding to the photoperiod of each light: dark (L: D) cycle (Buyse and Decuypere, 1988). However, feeding may occur during the dark period when birds are housed on a very short day length of 6 h (Morris, 1968). Murphy and Preston (1988) reported that broilers were 'restless rather than resting' under continuous light. Lack of sleep may reduce a bird's ability to cope with stressful conditions (Wiepkema, 1981).

Lighting Patterns and Body Composition

The published results about the effect of intermittent or increasing lighting programs on (abdominal) fat content are inconsistent. Intermittent lighting programs (Beane et al., 1979;

Ketelaars et al., 1986) and an increasing lighting program (Newcombe et al., 1992) were reported to increase body fat content of chickens as compared with a constant light. However, Deaton et al. (1978) and Weaver et al. (1982) found no significant difference in carcass ether extract or abdominal fat content between male broilers reared under 0.25L: 1.75D or 1L: 3D and a constant light. Charles et al. (1992) did not observe any change in body composition with an increasing lighting program either.

Malone et al. (1980), Cave (1981), and Cave et al. (1985), however, reported a reduction in abdominal fat content at slaughter of broiler chickens reared under an intermittent light program as compared with a constant light. Buyse et al. (1996a) found that both male and female broilers reared under a 1L: 3D intermittent light program had a lower abdominal fat content at 28 d of age compared with controls raised in a constant light. At 41 d of age, differences in abdominal fat content remained for male but not for female broiler chickens. Differences in endogenous (genotype and sex) as well as exogenous (duration timing, and severity of the short photoperiod schedule, dietary composition) factors in the above reports may explain these apparent discrepancies (Zubair and Leeson, 1996b).

The approach in the current experiments is that using a short lighting period for birds at an early age reduces feed intake and applies nutrient restriction. The early feed restriction results in retarded growth. When the lighting period is extended and the feed intake is restored to normal, the chicken is expected to exhibit compensatory growth in both body weight and adipose tissue. According to Wilson and Osbourn (1960), restricted nutrition at any stage of development had different retarding effects on different tissues: the earliest maturing tissues or parts being least, and the latest maturing ones (such adipose tissue) being most affected. Therefore, early mild feed restriction imposed by lighting programs was hypothesized to shift the onset and the full manifestation of the fat growth curve to a later age without affecting full body weight compensation, thus resulting in a leaner chicken at market age. Accompanying compensatory growth is an increase in feed efficiency.

As published results are inconsistent on the effect of lighting programs on body lipid content and growth performance, it is necessary to do more studies. The objective of the current study was to evaluate the effect of different lighting programs on broiler performance, body composition and mortality.

MATERIALS AND METHODS

Two experiments (Exp. 8.1 and 8.2) were conducted in the current study. In Exp. 8.1 and 8.2, the control light programs were so-called moderate lighting schedules (18L: 6D or 16L: 8D, Gordon, 1994), rather than traditional continuous or near continuous light. A 6-8 h dark period will allow the bird to have longer sleep and lower stress. The 6-8 h dark period can also allow the birds to empty its gut and force the bird to recycle nutrients from bone (Scott, personal communication), thus may result in stronger bones to support rapid growth (Classen and Riddle, 1989; Charles et al., 1992; Gordon, 1994).

Experiment 8.1

Exp. 8.1 was a factorial trial of 2 diets × 4 lighting programs. The effects of the two diets with different protein contents were reported in Exp. 4.2 in Chapter IV. Based on the literature, an increasing lighting program, an intermittent and an increasing-intermittent lighting program were developed and compared with the control light. All four lighting programs (Table 8.1) had 23-h light and 1-h dark during the first 3 d to let young chicks get used to the new environment. The control light (CON1) was 18L: 6D daily throughout 4 to 42 d. The increasing light (INC1) was 6L: 18D during 4 to 14 d, then, increased 4 h/wk until 18-h light/ 6-h dark at 29 d, which was maintained until 42 d. The intermittent lighting (INT) was 3L: 1D, then repeated throughout 4 to 42 d. The increasing-intermittent lighting (INC-INT) allowed photoperiod to increase from 1L: 3 D in every 4 h at 4 d to 3L: 1D at 29 d. The latter was maintained until 42 d. Both CON1 and INT had 771, while INC1 and INC-INT had 555 h of photoperiod during the entire growing period. The whole barn was divided into eight light-proof rooms. Each light treatment had two replicate lighting rooms and four replicate pens measuring 22 m^2 per pen. Each light room was equipped with two 100 W, 120 V incandescent light bulbs. Lighting intensity was 20 lx for the first 3 d and 5 lx from 4 to 42 d.

The experimental procedure is reported in Exp. 4.2 in Chapter IV. Briefly, a total of 2,400 male Peterson × Arbor Acre day-old chicks was randomly assigned to 16 floor pens in groups of 150 birds. The birds were raised to 42 d. Each lighting program had four replicate floor pens. Bulk weights of chickens in each pen were measured at 3 and 6 wk, respectively, and

Table 8.1 Lighting programs used in Exp. 8.1

Day	CON1[a]	INC1[b]	INT[c]	INC-INT[d]
0 to 3	23L: 1D	23L: 1D	23L: 1D	23L: 1D
4 to 14	18L: 6D	6L: 18D	3L: 1D	1L: 3D
15 to 21	18L: 6D	10L: 14D	3L: 1D	1.67L: 2.33D
Total light h, 0-3 wk	393	205	393	205
22 to 28	18L: 6D	14L: 10D	3L: 1D	2.33L: 1.67D
29 to 42	18L: 6D	18L: 6D	3L: 1D	3L: 1D
Total light h, 3-6 wk	378	350	378	350
Total light h, 0-6 wk	771	555	771	555

[a] Control lighting; [b] Increasing lighting; [c] Intermittent lighting; [d] Increasing-intermittent lighting program.

feed intake was recorded for 0-3 and 3-6 wk, respectively. Four chickens per pen, i.e., 16 chickens per lighting program were randomly collected at each of 2, 3, 5 and 6 wk of age for body composition analysis. All dead bnirds were necropsied. Death was attributed to SDS if there was no evidence of other disease and the birds were in good body condition with a full digestive tract, a small or empty gall bladder, and contracted heart ventricles (Blair, et al, 1991).

Experiment 8.2

This was a 3 diet × 4 lighting program × 2 sex factorial experimental design as Exp. 6.1 reported in Chapter VI. The three diets were based on corn, barley or wheat as the sole cereal respectively. Three increasing lighting programs (INC2, INC3 and INC4) were developed in Exp. 8.2 based on the results of Exp. 8.1. For the first 3 d, a near-continuous light (23L: 1D) was provided for all the chicks under each lighting program. The control light (CON2) was 16 L: 8 D for 4 to 42 d. INC2 was 6L: 18D, 4 to 9 d; 10L: 14D, 10 to 14 d; 14L: 10D, 15 to 21 d; 16L: 8D, 22 to 42 d. INC3 was 16L: 8D, 4 to 9 d; 6L: 18D, 10 to 14 d; 10L: 14D, 15 to 21 d; 14L: 10D, 22 to 28 d; 16L: 8D, 29 to 42 d. INC4 was 16L:8D, 4 to 9 d; 6L:18D, 10 to 14 d; 16L:8D, 15 to 21 d; 12L:12D, 22 to 35 d; 16L:8D, 36 to 42 d. The light intensity was 20 lx for 1 to 3 d and 5 lx for 4 to 42 d (see Table 8.2).

The experimental procedure was reported in Chapter VI. Briefly, a total of 2,880 day-old sexed broiler chicks (Peterson × Arbor Acres) was randomly assigned to 48 floor pens (either 60 males or females/pen). There were 12 floor pens for each lighting program. The birds were raised to 42 d of age. Body weight and feed intake were measured as in Chapter VI. Four birds each pen were randomly collected at each of 21 and 42 d of age for body composition analyses.

Table 8.2 Lighting programs in Exp. 8.2

Day	CON2[a]	INC2[b]	INC3[c]	INC4[d]
0 to 3	23L: 1D	23L: 1D	23L: 1D	23L: 1D
4 to 9	16L: 8D	6L: 18D	16L: 8D	16L: 8D
10 to 14	16L: 8D	10L: 14D	6L: 18D	6L: 18D
15 to 21	16L: 8D	14L: 10D	10L: 14D	16L: 8D
Total light h, 0-3 wk	357	253	265	307
22 to 28	16L: 8D	16L: 8D	14L: 10D	12L: 12D
29 to 35	16L: 8D	16L: 8D	16L: 8D	12L: 12D
36 to 42	16L: 8D	16L: 8D	16L: 8D	16L: 8D
Total light h, 3-6 wk	336	336	322	280
Total light h, 0-6 wk	693	589	587	587

[a] Control lighting 2; [b c d] Increasing lighting program 2, 3 and 4, respectively.

RESULTS

Experiment 8.1

No interaction between diets and lighting programs were observed for any of the parameters, therefore only the light effects were reported here. The two intermittent lighting schedules (INT and INC-INT) did not reduce the feed intake during 0-3 wk, thus did not affect body weight at 3 wk of age as compared with the control light (CON1). The feed: gain ratios for the birds reared under the two lighting programs were similar to that of the control birds ($p>0.05$). However, the increasing lighting program (INC1) significantly reduced the feed intake during 0-3 wk and body weight at 3 wk as compared with CON1 ($p<0.01$, Table 8.3). The feed: gain ratio for INC1 was similar to that for CON1 during 0-3 wk. During 3-6 wk, all the bird groups had similar feed intake, but the body weight at 6 wk was significantly lower for INC1 birds than for other lighting schedules ($p<0.05$, Table 8.3). Feed: gain ratio was similar among the lighting schedules during 3-6 wk. The body fat, body protein and body energy contents at 3 and 6 wk of age were similar among the four lighting programs (Table 8.4). There was no significant difference ($p>0.05$) in dietary energy and protein retention efficiencies during any phase among the light programs. The proportion of retained energy allocated to body fat was almost the same during 0-3 and 0-6 wk for the four lighting programs (Table 8.4). The effect of dietary protein level on body fat composition was reported in Chapter IV.

Table 8.3 Effect of lighting programs[1] on male bird performance in Exp. 8.1[2]

	Body weight[3], g		Feed Intake, g/d/bird			Feed: gain ratio, g/g		
	3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk
CON1	855[a]	2496[a]	53[b]	157	99	1.40	2.02	1.78
INC1	783[b]	2354[b]	48[c]	156	96	1.39	2.08	1.82
INT	860[a]	2465[a]	55[a]	159	100	1.43	2.07	1.82
INC-INT	858[a]	2513[a]	53[b]	162	100	1.39	2.06	1.80
	**	*	***					
SEM[4]	10	29	1	5	2	0.01	0.04	0.02
N[4]	4	4	4	4	4	4	4	4

[abc]Under the same column, means with different superscripts are significantly different, *: $p<0.05$; **: $p<0.01$; ***: $p<0.001$.

[1] CON1: control light; INC1: increasing lighting program; INT: intermittent lighting program; INC-INT: increasing-intermittent light program.

[2] Exp. 8.1 was a 4 lighting program × 2 diet factorial design with male broilers only. The two-way interaction between lighting program and diet was not significant for any parameter above ($p>0.05$).

[3] Mean of bulk weight of replicate pens.

[4] SEM: standard error of mean; N: number of observations.

Experiment 8.2

The three-way interaction of diet × light × sex was not significant ($p>0.05$). The results of the three diets and sex on performance and body composition were reported in Chapter VI. The three test lighting programs reduced body weight of broiler chickens at 3 wk of age by 9-13% as compared with CON1 ($p<0.05$, Table 8.5). The birds kept under INC2, INC3 and INC4 demonstrated compensatory growth during 3-6 wk. The body weight of the birds raised under the three test light programs at 6 wk of age was similar to that of birds raised under CON2, except that the birds kept under INC2 were just 4% lower than the control (CON2) birds ($p<0.05$, Table 8.5). Male birds raised under INC4 had higher ($p<0.05$) 3-wk body weight than male birds raised under INC3 (660 vs. 634 g/bird, not listed in the table). Females raised under the two lighting programs had similar 3-wk body weight, i.e., 604 vs. 603 g/bird, for INC4 and INC3, respectively.

The three test lighting programs reduced feed intake by 13-17% during 0-3 wk compared with CON2 (Table 8.5). The feed intake during 3-6 wk was similar among birds under the four lighting programs except that INC4 birds consumed 3% lower feed than CON2 birds ($p<0.05$,

Table 8.4 Effect of lighting programs[1] on body composition and energy retention efficiency in Exp. 8.1[2]

	Body fat content, %				Body protein[3], %		Body energy[3] kcal/kg		Energy retention[4]		Protein retention[5]		*Kf*[6]	
	2 wk	3 wk	5 wk	6 wk	3 wk	6 wk	3wk	6 wk	0-3wk	0-6wk	0-3wk	0-6wk	0-3	0-6wk
CON	8.6	9.9	14.4	15.6	17.3	18.2	1904	2408	0.46	0.50	0.51	0.53	0.49	0.57
INC1	8.7	9.7	14.1	15.2	16.5	19.1	1839	2414	0.47	0.51	0.52	0.56	0.50	0.56
INT	8.6	10.0	14.9	15.3	17.4	19.5	1936	2463	0.47	0.52	0.53	0.57	0.50	0.55
INC-INT	8.3	10.0	14.1	14.9	17.4	18.6	1932	2447	0.49	0.51	0.54	0.54	0.50	0.57
SEM[7]	0.4	0.3	0.4	0.5	0.4	0.4	31	52	0.02	0.02	0.02	0.01	0.01	0.01
N[7]	16	16	16	16	16	16	16	16	4	4	4	4	16	16

[1] CON1: control light; INC1: increasing lighting program; INT: intermittent lighting program; INC-INT: increasing-intermittent lighting program.

[2] Exp. 8.1 was a 4 lighting program × 2 diet factorial design with male broilers only. The two-way interaction between lighting program and diet was not significant for any parameter above ($p>0.05$).

[3] Body protein (%) =100 -water% -fat% -ash%; Body energy (kcal/kg) =body fat (g/kg) ×9.2 +body protein (g/kg) ×5.6, on fresh weight basis.

[4] Energy retention =average carcass energy/g of four birds from a pen/(AME/gain).

[5] Protein retention=[average carcass protein% of four birds from a pen × average body weight in a pen]/protein intake per bird.

[6] Partition coefficient of retained energy to fat (k_f)= energy retained as fat/total retained energy.

[7] SEM: standard error of mean; N: number of observations.

Table 8.5 Effect of lighting programs[1] and sex on the growth performance in Exp. 8.2[2]

	Body weight, g		Feed: gain ratio, g/g			Feed intake, g/d/bird			Mortality	SDS[3]
	3 wk	6 wk	0-3 wk	3-6 wk	0-6 wk	0-3 wk	3-6 wk	0-6 wk	%	%
CON2	692[a]	2201[a]	1.47a	1.90[a]	2.11[a]	46[a]	151[a]	96[a]	3.7	1.6[a]
INC2	602[d]	2115[b]	1.41[b]	1.88[a]	2.07[a]	38[c]	148[ab]	91[b]	2.2	0.4[ab]
INC3	619[c]	2147[ab]	1.42[b]	1.88[a]	2.08[a]	39[c]	150[a]	92[b]	3.1	1.2[ab]
INC4	630[b]	2161[ab]	1.43[b]	1.84[b]	2.01[b]	40[b]	146[b]	91[b]	3.9	0.1[b]
SEM[4]	3	15	0.01	0.01	0.02	0.0	0.0	0.0	0.0	0.0
Significance	*	*	*	*	*	*	*	*		*
N[4]	330	330	12	12	12	12	12	12	330	330
Light × sex[2]	*									

[abcd] Within a column, means with different superscrips are significantly different, * p≤0.05.

[1] CON2, INC2, INC3 and INC4 are control, increasing lighting program 2, 3 and 4 respectively.

[2] Exp. 8.2 was a 4 lighting program × 3 diet × sex factorial design. Other significant interactions (p<0.05) are reported in Chapter VI.

[3] SDS: sudden death syndrome;

[4] SEM: standard error of mean; N: number of observations.

Table 8.5). The feed: gain ratio was significantly lower (p<0.05) for the three test lighting programs during 0-3 wk, and remained significantly lower for INC4 (p<0.05) during 3-6 wk than for CON2 (Table 8.5).

Total mortality was not significantly affected by the lighting programs, but mortality from SDS was significantly lower for the birds reared under INC4 than for the birds kept under CON2 (Table 8.5).

Birds reared under INC2, INC3 and INC4 contained significantly higher water and lower fat (p<0.05, Table 8.6) at 3 wk, and higher water (p<0.05), higher protein (p<0.05) and lower fat (but p=0.11) at 6 wk of age than those reared under CON2 (Table 8.6). The body fat contents of the test light groups were about 12% lower at 3 wk and about 4-5% lower at 6 wk than that of the control birds. Carcass energy contents of the birds given the test lighting programs were all lower than that of birds given CON2, but the difference was not significant (p>0.05, Table 8.6).

The birds under the three test lighting programs were more efficient in utilization of dietary protein for body protein retention and less efficient in utilization of dietary energy for body energy retention (p<0.05) than the CON2 birds during 0-3 wk. The birds under the test lighting programs partitioned significantly (0-3 wk) less of retained energy to body fat deposition

Table 8.6. Effect of lighting programs[1] on carcass composition, and energy retention in Exp. 8.2[2]

	Carcass water, %		Carcass protein[3], %		Carcass fat[3], %		Carcass energy[3]	Protein retention[4]		Energy retention[5]		k_f[6]	
	3 wk	6 wk	3 wk	6 wk	3 wk	6 wk	6 wk	0-3 wk	0-6 wk	0-3 wk	0- 6 wk	0-3 wk	0-6 wk
CON2	69.9 ^{b}	64.2 ^{b}	18.3	19.2^{a}	9.6^{a}	14.0^{a}	2403	0.56^{b}	0.47^{a}	0.44^{a}	0.43	0.47^{a}	0.54
INC2	70.9 ^{a}	65.0^{ab}	18.2	19.0^{b}	8.4^{b}	13.4^{b}	2326	0.59^{a}	0.47^{b}	0.43^{ab}	0.42	0.43^{b}	0.54
INC3	71.1 ^{a}	65.3 ^{a}	18.2	18.9^{b}	8.5^{b}	13.6^{ab}	2295	0.59^{a}	0.47^{b}	0.42^{b}	0.42	0.43^{b}	0.54
INC4	71.0 ^{a}	65.0^{ab}	18.1	19.0^{b}	8.4^{b}	13.5^{b}	2313	0.59^{a}	0.49^{a}	0.42^{b}	0.42	0.43^{b}	0.54
SEM[7]	0.2	0.4	0.1	0.1	0.1	0.2	32	0.003	0.003	0.003	0.004	0.01	0.01
Significance	**	*		*	***	0.11		**	***	**		***	
N[7]	47	47	47	47	47	47	47	12	12	12	12	47	47

[abc] Under the same column, means with different superscripts are significantly different, * p≤0.05; ** P≤0.01, ***: p≤0.001.

[1] CON2: control lighting; INC2, INC3 and INC4: increasing lighting program 2, 3 and 4, respectively.

[2] Exp. 8.2 was a 4 lighting program × 3 diet × sex factorial design. Interactions between light and sex are not significant (p>0.05) for the above parameters. Other significant interactions are listed in Table 6.5 in Chapter VI.

[3] On fresh weight basis. Carcass energy was measured using bomb calorimeter; carcass protein (%) = 100 - water % - fat % - ash %.

[4] Protein retention=[average carcass protein% of four birds from a pen × average body weight in a pen]/protein intake per bird.

[5] Energy retention efficiency =average carcass energy/g of four birds from a pen/(AME/gain).

[6] Partition coefficient of stored energy to fat (k_f)= energy stored as fat/total retained energy.

[7] SEM: standard error of mean; N: number of observations.

than the CON2 birds (Table 8.6). Interactions between light and diet and between light and sex for all the parameters in Table 8.6 were not significant ($p>0.05$).

DISCUSSION

Lighting Programs, Feed Intake and Body Weight

Experiment 8.1

One of the prerequisites for compensatory growth is reduced growth at early age. However, both INT and INC-INT did not affect bird body weight at 3 wk of age compared with CON1, thus the two programs failed to achieve the goal of the current study.

The INC1 birds had lower feed intake thus lower weight gain than the CON1 birds due to a short photoperiod in INC1 compared with CON1 during 0-3 wk. This reduced body weight at an early age is the basis for compensatory growth in later age and for a delay of body fat accretion. During 3-6 wk, the birds reared under INC1 did not catch up with those reared under CON1 in body weight, indicating that no compensatory growth occurred in the growth-retarded birds under INC1. Yu et al. (1990) did not observe compensatory growth by market age with broilers subjected to early feed restriction. Our results in this experiment support the results of Yu et al. (1990).

Wilson and Osbourn (1960) pointed out that longer or more severe nutritional restriction may diminish the ability to recover from the effect of undernutrition. Most workers recommend feed restriction of not more than 7 and 5 days for male and female broilers, respectively, to allow for full body weight recovery (McMurtry et al., 1988; Plavnik and Hurwitz, 1991). In the current study, the birds kept under INC1 experienced 11 days of a 6L: 18D lighting schedule. The feed restriction imposed by the short photoperiod at early age was probably too long for the birds to recover their body weight at market age.

Another possible explanation for the absence of compensatory growth in the INC1 birds is that the two diets containing 2900-3000 kcal AME/kg used in Exp. 8.1 (see Table 4.1 in Chapter IV) might not have been high enough for the needs of compensatory growth. One of the two diets in Exp. 8.1 contained low protein (16.5%), as opposed to 18% recommended by NRC (1994), during 3-6 wk, which might also be too low for the compensatory growth of INC1 birds.

Wilson and Osbourn (1960) identified that the higher the plane of nutrients in diets upon re-alimentation, the more rapid and the greater the recovery in body weight of retarded animals. Plavnik and Hurwitz (1989) also observed higher requirements of broilers for most of the essential amino acids during the realimentation period, especially during the first 2 weeks of refeeding. In addition, the shorter light hours (about 30 hr shorter) in INC1 during 3-6 wk than in CON1 might be the third reason for the non-compensatory growth in the INC1 birds.

Experiment 8.2

The three test light programs (INC2, INC3 and INC4) were 104, 92 and 50 hrs shorter, respectively, than CON2 (357 h photoperiod) during 0-3 wk, leading the feed intake to be 17, 15 and 13% lower than that of CON2 birds, respectively, during 0-3 wk. The lower the feed intake, the lower the body weight. The three test lighting programs significantly reduced the feed/gain ratio during the first 3 wk as compared with CON2, indicating the birds with a lower feed intake utilized the nutrients more efficiently for body weight gain. The reduced feed intake, thus body weight during the first 3 wk for the test lighting programs posed a potential for a delay of fat deposition and body weight compensatory growth in late age.

By 6 wk of age, the birds raised under INC3 and INC 4 obtained complete compensatory growth in body weight, while the birds reared under INC2 were just 4% lower ($p<0.05$) than the control birds. INC2 birds did not completely catch up with the control birds in body weight, probably because the early feed restriction imposed by INC2 was too early and too long. The 6L: 18D schedule started at Day 4 in INC2 but at Day 10 in INC3 and INC4. INC2 was 104 h shorter, as opposed to 92 and 50 h shorter in photojperiod for INC3 and INC4 than CON2 during 0-3 wk.

In Exp. 8.2, total mortality was not affected by the lighting programs, but mortality from SDS was lower for INC2, INC3 and INC4 than for CON2 (0.4, 1.2, 0.1 vs. 1.6%). This result was consistent with those of Classen and Riddell (1989) and Blair et al. (1993). Classen and Riddell (1989) suggested that slower early growth rates might account for some of the in SDS mortality reduction and improvements in leg health. In addition, increased bird activity associated with the increasing lighting programs might also be responsible for the lower SDS incidence with the birds raised under INC2, INC3 and INC4 in Exp. 8.2 (Blair et al., 1993).

Lighting Programs and Body Composition

Rosebrough et al. (1986) and McMurtry et al. (1988) reported depression in the activities of enzymes associated with hepatic lipogenesis during undernutrition but, during refeeding, there was a dramatic increase in such enzyme activities. Fat cell size and number experience the same change and so does total body fat deposition. The critical point is to adjust the timing and duration of nutrition restriction through lighting programs so that it does not allow the nutrition-restricted birds to catch up to the *ad libitum* fed birds in body lipid deposition, but allow a full recovery of body weight by market age.

Experiment 8.1

In Exp. 8.1, INC1 significantly ($p<0.001$) reduce early feed intake and body weight as compared with CON1, but there was no significant difference in the body fat, water, protein and energy contents between INC1 birds and CON1 birds at 3 or 6 wk of age. This was probably because the early feed restriction through INC1 affected body weight gain and fat deposition to the same extent. The body weight gain and body fat deposition were parallel after the feed restriction was gradually removed by increasing the photoperiod. In addition, the greater variation in the fat content data may also be responsible for the non-significantly difference in body fat content at all ages between INC1 and CON1 birds.

Experiment 8.2

The three increasing lighting programs in Exp. 8.2 altered the body composition. The body protein and water contents of birds raised under INC2, INC3 and INC4 at 6 wk were significantly higher ($p<0.05$) than the birds raised under CON2. The body fat content of broilers raised under the three test lighting programs was 4-5% lower ($p=0.11$, $n=47$) compared with those kept under CON2. The non-significant difference in body fat content is probably due to higher variation of fat content data than body protein and water contents.

With the body composition change, energy partition, and energy and protein retention efficiencies were altered. Lower ratio (k_f) of body fat energy/total body energy, higher protein and lower energy retention efficiencies were observed for the birds reared under the three increasing programs as compared with those kept under CON2 for 0-3 wk of age. The results

indicated that the lighting programs (INC2, INC3 and INC4) were fairly successful in reducing body fat accretion and maintaining similar final body weight in broiler chickens as compared with CON2.

However, in Exp. 8.1 the body fat content of INC1 birds was similar to that of CON1 birds at all ages. Newcombe et al. (1992) even observed an increased body lipid content in chickens under an increasing lighting program as compared with a constant light. The difference is probably related to the nature, severity and duration of undernutrition, the age at the commencement of undernutrition, the degree and pattern of re-alimentation and genetic difference (Wilson and Osbourn, 1960; Buyse et al., 1996b; Zubair and Leeson, 1996a). It is more complicated to reduce body fat accretion and to keep similar final body weight at the same time than just to maintain similar final body weight through utilizing early nutrition restriction. Researchers have noted the factors affecting compensatory growth of body weight and body composition (Wilson and Osbourn, 1960; Zubair and Leeson, 1996a), but the relationship between body weight compensation and the contents of compensatory growth has not yet been well understood. Therefore, the quantity and quality of compensatory growth can not be entirely manipulated and it is not surprising that different results have been observed.

CONCLUSION

Lighting programs can affect bird activity, feeding behavior and health. Early feed restriction through lighting programs reduces feed intake and body weight gain, and may result in compensatory growth and possible reduction of body fat deposition. In Exp. 8.1, INC1 (but not INT and INC-INT) reduced early feed intake, but did not achieve compensatory growth due probably to a too severe feed restriction as compared with the CON1. In Exp. 8.2, INC2, INC3 and INC4 lighting programs reduced early feed intake and body weight gain, and also gave rise to complete or nearly complete compensatory growth. The body fat content of the birds reared under INC2, INC3 or INC4 was lower ($p<0.11$) than that of those reared under CON2. The birds reared under INC2, INC3 or INC4 also had lower ($p<0.05$) mortality due to SDS. This study demonstrates that it is possible to reduce body fat accretion of broiler chickens by certain lighting programs without affecting final body weight.

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CHAPTER IX

GENERAL DISCUSSION, CONCLUSION AND RECOMMENDATIONS

Poultry meat and egg products may be preferred in the near future because of the potential grain deficit facing the world and because broilers have the highest feed efficiencies (Brown, 1995). Reducing body lipid deposition in the chicken, thus improving feed utilization efficiency for meat production would further help deal with the potential grain crisis. Body fat in broiler chickens is highly heritable and highly variable. These two characteristics suggest that genetic selection, nutritional manipulation and environmental control may effectively modify body fat deposition and production efficiency of broiler chickens.

GENETIC SELECTION

Genetic selection can effectively alter both body size and composition. In two experiments, the effect of genotype on body composition was tested. In Exp. 3.1, two commercial strains, Peterson × Arbor Acre and Ross × Hy-line, exhibited similar body lipid and protein content as well as growth performance. On average, the birds in the two strains contained 15.9% lipids and 17.8% protein at 5 wk of age across strain, sex and diets. In Exp. 3.2, the three commercial strains from the breeder (Shaver Poultry Breeding Farm) also showed similar growth performance and body composition (e.g., 15.3% body lipid). Among the three pure strains from the breeder, Cornish 1, selected for low abdominal fat and high breast meat yield, contained lower body lipids than Cornish 2, selected for growth rate. Cornish 2 grew the fastest, deposited most body lipids, and exhibited the highest rate of leg abnormalities among the three pure strains. White Rock birds grew the most slowly but used feed more efficiently for weight gain. The results indicate that genetic selection is a very effective tool to change body size and composition in broilers. This study increased the available data on body composition of current commercial broiler chickens.

However, genetic selection for leanness may reach limits after several generations of genetic selection. Change of nutrition and the environment can further reduce the body lipid deposition of genetically lean broilers.

NUTRITIONAL MANIPULATION

There are a number of nutritional factors affecting broiler body lipid deposition. Dietary protein and amino acid contents and dietary cereal/fiber are the few factors investigated in the current study.

Dietary Protein Content

In two experiments, the high-protein (HP) diet significantly reduced body fat accretion and significantly increased body weight gain and feed efficiency as compared with the low-protein (LP) diet. The HP diet significantly decreased energy retained in the body as compared with the LP diet. With increased body energy retention per kg $W^{0.75}$, an increasing proportion of the total energy retained was directed to body lipids. However, a high-protein die will result in high nitrogen excretion to the environment and is a severe waste of precious protein resources. Therefore, producers may be required to use a low-protein diet with supplementation of limiting amino acids in the future to reduce environmental nitrogen pollution without losing performance of broilers.

Dietary Essential Amino Acids

A low-protein diet supplemented with synthetic AA was reported to increase body lipid deposition, although the diet substantially reduced nitrogen excretion without affecting bird performance in some studies (Parr and Summers, 1991; Moran, 1992). Improvement of dietary amino acid balance and increasing dietary essential amino acid content not only enhances body protein accretion, but also depresses fatty acid synthesis (Yeh and Leveille, 1969; Rosebrough et al., 1988; Grisoni et al., 1991).

Three low-protein diets with alternative amino acid profiles coupled with a 4-phase feeding program were tested in Exp. 5.1 to see if the low-protein diets would not increase body lipid deposition as compared with a control diet with a three-phase feeding program based on NRC (1994). The formulation of three test diets (T1, T2 and T3) was based on the amino acid profile

established according to maintenance amino acid requirements and carcass and feather amino acid profile plus the amino acid digestibility data. The three test diets contained 12-22% lower protein than the control diet. The four diets were compared using 1,440 male broiler chicks in a single-factor experiment. The results in this experiment indicated that the three test diets were successful to maintain growth performance and breast yield without significantly ($p>0.05$) increasing body lipid deposition. The three low protein diets also significantly ($p<0.05$) reduced excreta nitrogen content as compared with the control diet.

With the number of commercial amino acids increasing and the price decreasing, it is feasible and practical to use synthesized amino acids to enhance dietary essential amino acids and thus reduce dietary protein content. This is a very promising and effective approach to reverse the lipid-increasing effect associated with a reduced dietary protein level. Currently, it is important to conduct more quantitative research to explore, to what extent and under what essential amino acid content and balance, dietary protein can be reduced without affecting body lipid content.

Dietary Cereal/Fiber

Barley, wheat and/or their fiber fractions were reported to reduce liver lipid deposition and liver fatty acid synthesis in poultry as compared with corn (Maurice and Jensen, 1979; Chohchi et al., 1984). However, the effect of dietary cereal/fiber on body lipid deposition in broiler chickens is not available in the published literature to the author's best knowledge.

In two experiments, it was observed that both barley and wheat diets significantly reduce bird body fat accretion at 6 wk of age as compared with a corn diet without affecting final body weight. The higher the hulled barley or wheat inclusion percentage in the diet, the lower the body lipid content. It was shown that wheat bran (but not barley spent grain) addition to a corn-based diet reduced body fat deposition compared with the corn alone diet.

It was further confirmed that the barley and the wheat diets significantly reduced bird body fat accretion compared with the corn diet. The activities of key enzymes (FAS and ACC) in liver fatty acid synthesis were consistent with the corresponding body fat content. Incorporation of labelled water [^3H_2O] into liver fatty acids had a pattern consistent with that of the key enzyme activities. It was concluded that barley or wheat in a diet reduced liver fatty acid synthesis,

contributing to lower body fat content in broiler chickens fed barley or wheat based diets. Dietary β-glucans and xylans do not seem to affect lipid metabolism in broiler chickens.

These are important findings, but are just some initial work in this area. There are still a lot of questions to be answered and also the current results need to be confirmed by researchers in other laboratories. It may be necessary to further study the effect of the components in barley and wheat on the body lipid deposition and liver fatty acid synthesis.

ENVIRONMENTAL FACTORS

Lighting patterns in a light-proof house affect feed intake, activity, health and well being of broiler chickens thus changing growth and lipid deposition patterns. Early feed restriction by a short photoperiod results in retarded growth. The subsequent extent of compensatory growth in body size and lipid content depends on several factors, such as the duration, timing, nature and severity of undernutrition, the condition of re-alimentation, and genetic difference, e.g., sex and strain (Zubair and Leeson, 1996). Currently, little is understood about the extent of the above factors in order to control body lipid deposition without affecting final body size by feed restriction.

In two experiments (Exp. 8.1 and 8.2), the effect of lighting programs on lipid deposition and performance was tested in broiler chickens. There was no compensatory growth in body weight with an increasing, an intermittent and an increasing-intermittent lighting program compared with a control (18L: 6D) in Exp. 8.1. However, complete compensatory growth in body size but not in body lipids was achieved with two increasing lighting programs (INC2, INC3 and INC4) as compared with the control light (16L: 8D, CON2) in Exp. 8.2. A possible explanation is that the feed restriction by INC1 in Exp. 8.1 was too early and too severe for the birds to recover the body weight to the normal size after the lighting period was extended and the feed intake was restored to normal. The incidence of SDS mortality for INC2, INC3 and INC4 birds was much lower than for CON2 birds, but only significantly lower for INC4 birds. The results indicate that it is possible to lower body lipid accretion without affecting final body weight through changing lighting pattern.

Unlike early severe feed restriction in which birds do not grow during the feed restriction period, early feed restriction imposed by lighting programs is natural, mild and acceptable by

animal welfare standards and by poultry producers. The INC2, INC3 and INC4 lighting programs are recommended for use in commercial poultry production. In the future, additional research is needed to study effect of duration, timing, and the severity of the short photoperiod in an increasing lighting program and timing and pattern to restore the lighting regime to normal.

OVERALL CONCLUSION

According to the results in the current study, it is recommended that the broiler producer obtain a relatively lean strain of commercial broiler chicks, and feed the birds a diet composed of barley and wheat as the main cereals during 3-6 wk of age. The dietary protein level can be formulated to be 21 and 18% during 0-3 and 3-6 wk of age, respectively, and the amino acid profile can be similar to that of Diet T1. The lighting program can be an increasing lighting schedule as the one in Exp. 8.2. Adoption of the above feeding and/or lighting programs will result in leaner chickens without affecting body weight gain and should give a reduction in nitrogen excretion, metabolic diseases and mortality. Although it is not clear if the above programs have additive effect, these programs may not conflict with each other in terms of reducing body lipid deposition based on the current knowledge. This suggested program is sustainable, environmentally friendly and beneficial.

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APPENDIX I

EVALUATION OF AN EM-SCANNER TO PREDICT BODY FAT IN BROILER CHICKENS

ABSTRACT

The total body electric conductivity (TOBEC) of a live bird in an Electro-Magnetic Scanner (EM-Scanner) is proportional to its free electrolyte concentration, and may be used to accurately estimate fat-free mass (FFM). It was considered it might be possible to calibrate the EM-Scanner to predict body fat content, as fat mass + fat-free mass = body weight, and fat mass/body weight = fat content. In five experiments, the EM-Scanner was tested. Samples of live chickens were scanned in a chamber mounted to a base detector unit to obtain TOBEC. Later all the chicken samples were analyzed for water, fat, and ash, and fat mass and fat-free mass were calculated. Various regression and correlation analyses between FFM, fat mass, total body water or fat content and TOBEC (or/and body weight) were conducted. The results of the five experiments indicated that FFM can be accurately predicted from TOBEC (E) using this equation FFM = a + b E (a is intercept and b is slope, R^2 = 0.81-0.97). Fat content was not directly correlated with TOBEC, but could be indirectly predicted using this equation Fat (%) = [body weight - (a + b E)]/body weight × 100. However, the predicted fat content differed greatly from the measured fat content. Furthermore, FFM could be more accurately predicted from body weight itself than from TOBEC. Therefore, the approach fat (%) = [body weight - (a + b E)]/body weight × 100 was totally unnecessary. It was concluded that the EM-Scanner was no better than a balance in respect of predicting FFM thus fat content of broiler chickens.

Key words: EM-Scanner, TOBEC, body fat, broiler chicken

INTRODUCTION

The most accurate method used to quantify body composition is chemical analysis of a homogeneous body sample. But this method is very time-consuming and expensive. Also, it can not be used in live animals. Some indirect methods are available, but each has its own advantages and limitations.

Bio-electrical impedance analysis (BIA) is a widely used method for estimating body fat-free mass in humans (National Institutes of Health, 1994; Baumgartner, 1996). A similar equipment to BIA is the Electro-Magnetic Scanner (EM-Scanner). The underlying principle in the EM-Scanner is the same as in BIA: the conductivity of a tissue is dependent on its water and free electrolyte concentration, temperature, and the frequency of the current (Baumgartner, 1996). Lean tissues (with the exception of bone) have about 20 times the conductivity of fat (Geddes and Baker, 1967; Pethig, 1979). Thus, total body electrical conductivity (TOBEC) is directly proportional to the volume of the fat- and bone-free body mass. The EM-Scanner was considered to be a highly reliable and relatively accurate means of estimating total body water and fat-free mass of a live subject (Baumgartner, 1996). The approach to measurement in the EM-Scanner is completely different from that in BIA. In the EM-Scanner, an electric current is induced in the body using an electro-magnetic field without physical contact. This is accomplished by inserting the body into a large electric coil, or solenoid, that generates an electro-magnetic field (Baumgartner, 1996). A major advantage of the technique is that repeated measurements of body fat-free mass or total body water can be rapidly and easily obtained on live subjects with minimal discomfort to the animal.

It was considered that the EM-Scanner might also be used to estimate body fat, as fat mass + fat-free mass = body weight, and fat mass/body weight = fat content. The fat content of birds might be predicted from body weight and TOBEC using the following equation if fat-free mass (FFM) can be accurately predicted:

$$\text{Fat (\%)} = (\text{body weight} - \text{fat-free mass})/\text{body weight} \times 100$$

Application of this technique to poultry species may provide a new tool to monitor changes and difference in broiler body composition.

MATERIALS AND METHODS

Measurement of Chicken TOBEC

Sample chickens were randomly collected at various ages in each of Exp. 3.1, 3.2, 4.2, 5.1 and 6.1, weighed and scanned in the EM-Scanner SA-3000 Small Animal Body Composition Analyser (Meat Quality Inc., Springfield, IL 62702). The operation of the EM-Scanner was as follows. A live bird was put into an elastic nylon bag to a football shape, then inserted with a carrier to a right-size chamber mounted to a base detector unit. The EM-Scanner SA 3000 had 3 chambers of different sizes. Chamber 3114, 3152 and 3203 were 114, 152 and 203 mm, respectively, in diameter of the chamber. Finally, TOBEC (E) was recorded. Each bird was scanned live 2 to 3 times to get consistent readings. Then, the bird was taken out from the chamber and killed by cervical dislocation. The head and neckwere removed, also wings and legs before the carcass was bagged and frozen, as these parts might not be well-cooked in the autoclave oven, and might generate error in sub-sampling. Thus body weight (WT) and TOBEC (E) were obtained from live birds; and fat content, water content, FM, FFM and TBW were obtained from corresponding carcasses with those parts removed. In Exp. 3.1 and 4.2, body length of the bird in the nylon bag was also measured.

Laboratory Analysis

Later, all the frozen chicken samples were cooked in the autoclave oven for 6 to 14 hours depending on size, blended, sub-sampled, and freeze-dried. Body water, ether extract, and ash content of each bird sample were analyzed in the laboratory (see Exp. 3.1). The data of FM, FFM, and TBW were then obtained. Fat mass (FM) = carcass fat content (%) × body weight. Fat-free mass (FFM) = total body weight - fat mass. Total body water (TBW) = carcass water content (%) × body weight. Later correlation and regression analysis were conducted using the two sets of data.

Table A1 Composition of simulated chicken standards

Standard[1]	Water, g	Chicken meal, g	Tallow, g	Total, g
1	1300	700	0	2000
2	1300	540	160	2000
3	1300	350	350	2000
4	1300	180	520	2000
5	1100	900	0	2000
6	1200	727	73	2000
7	1300	545	155	2000
8	1400	364	236	2000
9	1440	810	0	2250
10	1280	720	0	2000
11	1120	630	0	1750
12	960	540	0	1500

[1] Standard 1- 4: weighing 2000 g each and containing a constant water (65%) and increasing fat contents.
Standard 5-8: weighing 2000 g and containing constant fat (21%) and increasing water contents.
Standard 9-12: weight decreasing and containing the same water, fat, and protein contents.

Table A2 Standard sodium chloride solutions

NaCl Standard	NaCl %, W/V	NaCl, g	Volume, ml
0	0.00	0.0	2000
1	0.05	1.0	2000
2	0.10	2.0	2000
3	0.15	3.0	2000
4	0.10	1.5	1500
5	0.10	2.4	2400

Simulating Standard

To test the relationship between TOBEC and fat percentage, FFM, water content, weight, and ion concentration of a given subject, 12 simulated chicken standards were designed and formulated from mixed dry chicken meal, tallow, and deionized water. The dry chicken carcass meal saved from the carcass samples contained dry matter 95.3%, fat 44.3%, and protein 49.0%. In addition, six NaCl solutions with different concentrations and volumes were made of deionized water and NaCl. All the standards were accommodated to the same shape in plastic bags of the same size. The composition of the 12 chicken standards and 6 NaCl solutions is listed in Table A1 and A2 respectively. The conductivity of the standards was measured using the large chamber (Chamber 3203). In addition, the conductivity of the deionized water, the dry chicken meal and the tallow were measured as well.

Correlation Analyses and Calibration of Regression Equations

In experimental barns, a set of data including body weight, body length in a nylon bag and TOBEC (E) of each bird were obtained. In the laboratory, another set of data including body water content, fat content, TBW, FFM and FM of each carcass were obtained. All varieties of correlation relationships were analyzed and regression equations were developed using the two sets of data and the linear model FFM = a + bE. Correlation and regression relationships of the data from the standard test were also analyzed using SAS program (SAS, 1996). The regression equations for body fat (%) and FM prediction developed in one trial were applied in the other to test the accuracy of the prediction.

Table A3 Correlation coefficients[1] between parameters (Exp. 5.1)

	WT[2] (g)	E[3]	Water[4] (%)	Fat[4] (%)	TBW[5] (g)	FFM[5] (g)	FM[5] (g)
WT (g)		0.892	-0.176	0.307	0.958	0.960	0.705
E	0.929		0.089	0.090	0.910	0.900	0.507
Water (%)	0.153	0.306		-0.903	0.011	-0.017	-0.737
Fat (%)	-0.081	-0.258	-0.846		0.143	0.143	0.881
TBW (g)	0.972	0.932	0.338	-0.218		0.997	0.591
FFM (g)	0.975	0.940	0.307	-0.252	0.993		0.590
FM (g)	0.524	0.334	-0.636	0.796	0.398	0.373	

[1] The correlation coefficients on the upper right side and lower left side are from birds at 2 and 7 wk of age respectively.
[2] WT -- live body weight, g.
[3] E -- TOBEC values for live birds.
[4] Water, Fat -- measured water (%) and fat content (%) of the carcass, respectively.
[5] TBW, FFM and FM -- measured total body water (g), fat-free mass (g) and fat mass (g) in the carcass.

RESULTS

Correlation Analyses

A correlation analysis between all relevant parameters was conducted in each of the five experiments. The correlation analysis results from the data in Exp. 5.1 were listed in Table A3. Table A3 indicated that fat content was poorly correlated directly with TOBEC (r= 0.09 and -

0.26 respectively for 2- and 5-wk-old birds). Fat mass was more highly correlated with TOBEC (r=0.54 and 0.33 respectively for 2- and 5-wk-old birds) than fat content. Fat-free mass (FFM) and total body water (TBW) were highly correlated with TOBEC (E) (r between 0.90 and 0.94). However, FFM or TBW was even more highly correlated with live body weight (WT) (r was between 0.96 and 0.98).

Table A4 Regression equations developed in TOBEC® calibration[4]

Trial	Dependent variable[1]	N	Sex[2]	Age, d	Chamber size[3]	Equation	R^2
Exp. 3.1	FFM	220	M, F	21	3114	270 + 0.328 E	0.837
	FFM	32	M, F	35	3152	466 + 0.676 E	0.932
	FFM	191	M, F	38	3203	534 + 1.180 E	0.895
Exp. 3.2	FFM	190	M, F	21	3114	213 + 0.396 E	0.863
	FFM	190	M, F	42	3203	666 + 1.101 E	0.912
Exp. 4.2	FFM	125	M	14-21	3114	221 + 0.538 E	0.968
	FFM	119	M	35-42	3152	794 + 0.964 E	0.963
	FFM	92	M	35-42	3152	749 + 0.780 E	0.927
Exp. 5.1	FFM	47	M	14	3114	151 + 0.466 E	0.811
	FFM	45	M	42	3203	775 + 1.050 E	0.916
	FFM	42	M	49	3203	977 + 0.967 E	0.883
Exp. 6.1	FFM	235	M, F	21	3114	247 + 0.350 E	0.939
	FFM	228	M, F	42	3203	688 + 1.075 E	0.947

[1] FFM--fat-free mass, g; TBW--total body water, g; FM--fat mass, g; E--TOBEC values; WT--live body weight, g

[2] M: male; F: female.

[3] Chamber size 3114, 3152 and 3203 are 114, 152 and 203 mm respectively in diameter of the chamber.

[4] WT and E were obtained from live birds, and FFM, TBW and FM were obtained form the carcasses with head and neck, legs and wings removed.

Regression Equation Development

As FFM was highly correlated with TOBEC (E), the following linear regression equation was developed to predict FFM:

$$FFM\ (g) = a + b\ E \qquad (1)$$

Where a is the intercept, and b is the slope. In each of the five experiments, similar regression equations were developed (R^2 = 0.811-0.968, Table A4). The intercepts or the slopes obtained in one experiment were different from those obtained in the other (Table A4). For example, for

Chamber 3114, the slopes were from 0.328 to 0.466. The larger the chamber size, the higher the slope. Body length basically did not affect FFM.

Accordingly, FM and fat content (%) were predicted using these two equations, respectively, obtained from data in a previous experiment:

$$FM\ (g) = WT - (a + b\ E) \quad (2)$$

$$Fat\ (\%) = [WT - (a + b\ E)]/WT \times 100 \quad (3)$$

However, the predicted FM or fat (%) differed greatly from the measured FM or fat content (not listed here).

Table A5. The simulated chicken standards and their TOBEC

Standard	1	2	3	4	5	6	7	8	9	10	11	12
Total weight, g	2000	2000	2000	2000	2000	2000	2000	2000	2250	2000	1750	1500
FFM[1], g	1700	1600	1500	1400	1600	1600	1600	1600	1913	1700	1488	1275
FM[2], g	300	400	500	600	400	400	400	400	87	300	512	725
Fat %	15	20	25	30	20	20	20	20	15	15	15	15
TBW[3], g	1300	1300	1300	1300	1100	1200	1300	1400	1440	1280	1120	960
Water %	65	65	65	65	55	60	65	70	65	65	65	65
Chicken meal, g	700	545	350	180	900	727	545	364	810	720	630	540
TOBEC[4]	1054	938	650	234	1252	1153	938	873	1439	1389	1046	773

[1] FFM--fat-free mass (g); [2] FM--fat mass (g); [3] TBW--total body water, g; [4] TOBEC-total body electrical conductivity.

Standards

The results of the standards in Table A5 indicated that (1) the TOBEC showed no relationship with fat content and fat mass (Standard 1-4 and 9-12). (2) TOBEC did not change in parallel with fat-free mass (FFM), which was indicated by Standards 5-8. (3) TOBEC was not

Table A6 The concentration of sodium chloride (NaCl) solutions and their TOBEC

NaCl Standard	NaCl, g/100 ml	Volume, ml	NaCl, g	TOBEC
0	0.00	2000	0	23
1	0.05	2000	1	226
2	0.10	2000	2	411
3	0.15	2000	3	549
4	0.10	1500	1.5	271
5	0.10	2400	2.4	454

related to either water content or water mass (Standard 1-4). (4) TOBEC was highly correlated with the dried chicken meal amount (Standard 1-12): the correlation between chicken meal (g) and TOBEC was 0.92. (5) Total weight (g) was poorly correlated with TOBEC (Standards 1-8). (6) With similar chicken meal amount (Standards 7 and 12, or 6 and 10), higher water amount or lower FM slightly increased TOBEC values.

The results in Table A6 indicated that the TOBEC value was proportional to the absolute amount of NaCl, instead of to the NaCl ion concentration or the solution volume. The correlation coefficient between NaCl (g) and TOBEC was 0.995.

DISCUSSION

Correlation and Regression Relationship

Correlation analysis (Table A3) indicated that fat content could not be directly predicted from body TOBEC since the direct correlation between fat (%) and TOBEC was very low ($r = -0.26$ to $+ 0.17$). Baumgartner (1996) also indicated that. However, FFM was highly correlated with TOBEC. This supports the findings of Staudinger et al., (1995) who also obtained a high correlation between TOBEC and FFM. Using an indirect method, fat (%) was predicted with this equation: fat (%) = (body wt – FFM)/body weight × 100% (here FFM was predicted using FFM (g) = a + b E obtained from a previous trial). But the predicted fat content was too far from the determined content. One possible reason is that the slope and intercept obtained in one trial to predict FFM were different from those obtained in the other. The different intercepts and slopes in Table A3 were related to room temperature (Meat Quality Inc., 1994; Baumgarter, 1996) at which TOBEC were measured, TOBEC range (i.e., chicken size range), and the number of birds measured (Baumgarter, 1996). The wider the range of the independent variable (i.e., TOBEC values), the more consistent the slopes and intercepts (Guo and Chumlea, 1996). The other possible reason is that FM is a small portion compared with FFM. A minor error in prediction of FFM may result in large deviation of the calculated FM since FM = body weight – FFM. It can be explained in this example. Say, a 2000 g of chicken has 1700 g FFM and 300 g FM. The fat content is 15%. If there is a 50-g difference in prediction of FFM, this represents an error only of 50/1700, i.e. 2.9%. As FM = body weight - FFM, FM also had 50 g difference,

but the 50 g difference for FM is an error of 50/300, i.e. 17%. Therefore, this approach is not feasible. There have been, actually, no published reports to predict body fat content using TOBEC in broilers, although the TOBEC® company declared that the EM-Scanner could be used to predict chicken fat content.

In the correlation analysis (Table A3), it was indicated that FFM was more highly correlated with body weight than with TOBEC. Thus, it would be more accurate to predict FFM from body weight directly. Therefore, it is absolutely unnecessary to use an EM-Scanner to measure TOBEC to predict FFM, then to calculate body fat content using the equation: fat (%) = (body weight – FFM)/body weight × 100%. From this point of view, an EM-Scanner is no better than a weight scale to predict body fat content.

Simulated Standard

In the modeled standards, it was found that the TOBEC values were actually poorly correlated with fat content, fat mass, fat-free mass, body water and total weight. The TOBEC values were only correlated with the content of dried chicken meal with a correlation coefficient of 0.915. Obviously, the TOBEC was just related to the ion amount in the chicken powder. This was confirmed in the sodium chloride solution test. This test indicated that the TOBEC values are highly correlated to the absolute amount of NaCl ion but not to the NaCl solution concentration or volume.

CONCLUSION

The Electro-Magnetic Scanner (EM-Scanner) can not be directly used to predict body fat content. The indirect method through estimating fat-free mass from TOBEC, then calculating fat content did not give a good estimation of body fat content of broiler chickens. Furthermore, since FFM was more highly correlated with body weight than with TOBEC, a better estimate of body fat content would be obtained if body weight was directly used to estimate fat-free mass, then calculate fat content. From this point of view, it was concluded that the EM-Scanner is no better than a balance.