

**Modelling Responses of Broiler Chickens
to Dietary Balanced Protein**

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to Dietary Balanced Protein**

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ABSTRACT

Protein is an important nutrient for growing broiler chickens, as it affects broiler performance, feed cost as well as nitrogen excretion. The objective of this dissertation was to develop a growth model for broiler chickens that could be easily used by practical nutritionists. The model should facilitate the selection of feeding strategies (in terms of dietary protein and energy) that results in the desired body composition of broilers while minimizing costs.

Two important theories that are generally used in animal growth models were validated for broiler chickens. It was confirmed that, where protein is limiting, protein deposition rate will not increase with additional energy intake. The second theory, stating that the fat-free body composition is independent from nutrition, is not a valid assumption for broiler growth models.

It was demonstrated that broiler responses to dietary balanced protein level (DBP) depend on previous protein nutrition. These results suggest that DBP levels in grower and finisher diets should not be optimised independently, but simultaneously.

A model was developed that predicts broiler responses (growth rate, feed conversion ratio, carcass yield and breast meat yield) to DBP level. The model makes it possible to construct tailor-made dose-response curves without actual experimentation.

Based on the predictions by this new model and data on feed and meat prices, the economic aspects of DBP level in broiler diets were evaluated. It was shown that formulating diets for maximum profit instead of maximum broiler performance may strongly increase profitability of a broiler production enterprise. Model simulations revealed as well that DBP level for maximum profitability depend on how the broilers are marketed; as whole birds, carcass or cut up.

Voor Lianne

Voor Jan-Jaap, Wilmer en Marilyn

Ter herinnering aan mijn vader

VOORWOORD

Een promotieonderzoek is in de eerste plaats een leerervaring. Onderzoek doen is immers een vak dat je kunt leren. Kiezen van de juiste doelen, ontwerpen, uitvoeren en analyseren van proeven en desk-studies, schrijven van wetenschappelijke artikelen, presenteren van resultaten op congressen, etc. Daarnaast had dit promotie-project een extra dimensie: het onderzoek is uitgevoerd voor en binnen een bedrijf. Logisch dus dat het project kennis moest opleveren die praktisch relevant en toepasbaar is. De voortdurende implementaties in de praktijk maakten het onderzoeksproject extra leuk en leerzaam, maar ook extra complex. Gelukkig heb ik het niet alleen hoeven doen. Naast dank aan God die mij de gaven en gezondheid gaf, wil ik graag de mensen bedanken die hun bijdrage hebben geleverd, direct of indirect.

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General Introduction

PROTEIN NUTRITION OF BROILER CHICKENS

The poultry meat industry in the 21st century has evolved from tens of thousands of small farms in the post-World War II period to an industry of relatively few large vertically integrated companies. World chicken meat production has grown from 29 million metric tons in 1990 to an estimated 50 million metric tons in 2000 (Aho, 2002). Nowadays, at least two kg of broiler feed is needed for each kg of chicken meat that is produced. As a result, a 1% improvement in efficiency of feed utilization for meat deposition in broilers would save at least 1 million metric tons of broiler feed on a worldwide scale.

Feed utilization in broilers is improved with higher protein levels in the diet. Dietary protein level also influences growth rate and body composition (carcass and breast meat yield) in broilers (Smith *et al.*, 1998). Moreover, the amount of protein in the diet affects feed cost as well as nitrogen excretion that may result in pollution of the environment. The mentioned effects of dietary protein in broiler diets depend on many interacting factors, like dietary energy level, broiler genotype and temperature (NRC, 1994). Even if knowledge is available of all these interactions, choosing the optimal protein level for a broiler diet remains a complex task for nutritionists. A growth simulation model that evaluates all the relevant interacting factors simultaneously, is needed for this aim (Gous, 1998).

BROILER GROWTH MODELS

Several models are available to simulate the growth of broiler chickens (Zoons *et al.*, 1991). Growth models can be classified as empirical and mechanistic models. Both types of models give a description of the relationship between a dependent variable and an independent variable. However, empirical models work as a black box whereas mechanistic models describe underlying biological processes (Zoons *et al.*, 1991).

For predicting the response of broilers to a certain diet, mechanistic models are preferable to empirical models. Empirical models are only reliable for the exact (research) conditions under which the model is constructed. Mechanistic models have a better chance to be valid for

extrapolation than empirical models, provided that the mechanisms of growth are represented in a correct way.

An example of a mechanistic broiler growth model is the Edinburgh Growth Model (EFG-model; Emmans, 1981; Emmans and Fisher, 1986; Gous, 1998). The EFG-model is unique in that it is based on a relatively clear, but at the same time rather complete growth concept, including a description of the genetic potential of a broiler (Eits, 1996). Given the ongoing and dramatic genetic development of broilers (Albers, 1998), growth models that do not deal explicitly with the effect of genetic potential will probably be outdated within several years. Another strong feature of the EFG-model is that it takes into account the relationship between ambient temperature and metabolism (Emmans, 1981). For those reasons, we believe that the EFG-model is more fit as a practical tool for broiler nutritionists than other broiler models mentioned by Zoons *et al.* (1991) or published more recently (King, 2001).

A problem with the practical use of the EFG-model is that it requires data on the protein mass in adult broilers to determine the potential growth curve. It is hardly possible to obtain representative data on adult broilers of a commercial breed (Zoons *et al.*, 1991). The difficulties with determination of the potential growth curve of a given genotype may explain the systematic differences in protein deposition rates between predictions by the EFG-model and unpublished data from trials in our laboratory (Eits, 1996). The observed bias could also mean, however, that the underlying nutritional theories are not completely valid for broiler chickens. An important theory in the EFG-model as well as in several growth models for pigs (Whittemore and Fawcett, 1976; Moughan and Versteegen, 1988) is that dietary protein will be preferentially used for protein deposition, unless dietary energy or other factors become limiting. The key assumption is that, where protein intake is limiting, protein deposition rate will not increase with additional energy intake. Experimental evidence that is relevant to the testing of this theory in broiler chickens is scarce.

According to another theory in the EFG-model (Emmans, 1981), water and ash deposition in the body is associated with protein deposition only (Black *et al.*, 1986; Moughan *et al.*, 1987). This implies that the relationships between water and protein mass, and between ash and protein mass, are both considered to be independent of nutrition. Opposing views exist on whether the fat-free body composition is indeed independent of nutrition

(Kyriazakis *et al.*, 1991; De Greef *et al.*, 1992). Moreover, this latter theory is hardly validated for broiler chickens.

Thus, for a successful application of models in practice, the required model input should be easily available for the user. Moreover, the underlying theories should be as simple as possible and sufficiently validated in order to gain user confidence. It is concluded that existing broiler growth models in their present form do not meet these criteria. Consequently, a practical tool for determining optimal nutrient levels in broiler diets is not available yet.

The objective of this dissertation was to develop a growth model for broiler chickens that could be easily used by practical nutritionists. The model should facilitate the selection of feeding strategies (in terms of dietary protein and energy) that results in the desired body composition of broilers while minimizing cost. A description of the genotype of a broiler chicken should be an input variable for the model.

OUTLINE OF THE DISSERTATION

It was mentioned as a key assumption in many growth models that dietary protein will be preferentially used for protein deposition, unless dietary energy or other factors become limiting. The validity of this theory for growing broiler chickens was tested in an experiment that is described in Chapter 2. Data of this experiment were also used to validate the other mentioned assumption in growth models, namely that fat-free body composition is independent of nutrition (Chapter 3). In Chapter 4, an experiment was described on the effects of early life protein nutrition on responses of broiler chickens to dietary protein at later age. Data of this latter experiment and from other experiments were used to develop a new model. The new model predicts broiler responses (growth rate, feed conversion ratio, carcass yield and breast meat yield) to dietary balanced protein level (Chapter 5). Based on the predictions by this model and data on feed and meat prices, the economic aspects of dietary protein level in broiler diets were evaluated (Chapter 6). In the General Discussion, the contribution of the studies in Chapters 2 to 6 to the realisation of the general objective of this dissertation is evaluated.

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Chapter 1

**Protein and Lipid Deposition Rates in Male Broiler
Chickens: Separate Responses to Amino Acids and
Protein-Free Energy**

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and K. H. De Greef, 2002. *Poult. Sci.* 81:472-480. Reproduced with
permission of The Poultry Science Association, Inc.

ABSTRACT Two experiments of similar design were conducted with male broiler chickens over two body weight ranges, 200 to 800 g in Experiment 1 and 800 to 1,600 g in Experiment 2. The data were used to test the hypothesis that protein deposition rate increases (linearly) with increasing amino acid intake, until energy intake becomes limiting for protein deposition rate. Additional amino acid intake above this point would be deposited less efficiently. An increase in energy intake would increase lipid deposition rate, but should, at low amino acid intakes, not affect protein deposition rate. Each experiment consisted of 18 treatments: two levels of protein-free energy ($\text{energy}_{\text{pf}}$) intake, combined with nine amino acid to $\text{energy}_{\text{pf}}$ ratios. Protein was balanced for amino acid content and lysine was the first limiting amino acid in the diet.

Protein deposition rate increased with additional amino acid intake. No evidence was found that $\text{energy}_{\text{pf}}$ intake limited protein deposition rate at high amino acid intake. Extra intake of $\text{energy}_{\text{pf}}$ increased lipid deposition rate, which was independent of amino acid intake. Where amino acid intake was limiting, additional intake of $\text{energy}_{\text{pf}}$ had generally no effect on protein deposition rate. The marginal efficiency of amino acid utilization for protein deposition did not depend on body weight. The facts are relevant to the modelling of the growth of broiler chickens.

INTRODUCTION

The rate at which broiler chickens grow, and the composition of the growth, is determined in large part by their intakes of amino acids and energy. The major costs of feeding are those of supplying amino acids and energy. It follows that amino acids and energy are the most important nutrients to be considered in the design of feeding strategies. To do this, some way of predicting the responses of growth rate and body composition to these nutrients is needed.

Whittemore and Fawcett (1976) proposed that dietary protein will be preferentially used for protein deposition, unless energy availability or other factors (genotype or environment) become limiting. Moughan and Verstegen (1988) used the same idea in their model. The key assumption is that, where protein intake is limiting, protein deposition rate will not increase with additional energy intake. The idea has experimental support in pigs (Campbell and Taverner, 1988; Bikker, 1994) but seems less valid for pre-ruminant calves (Gerrits *et al.*, 1996). To the authors' knowledge, there is hardly any experimental evidence that is relevant to the testing of the idea in broiler chickens. The objective of this study was to investigate the separate effects of amino acid and energy intakes on the rates of protein and lipid deposition in the different body components of young broiler chickens. The main purpose was to test whether, at amino acid intakes that were limiting, energy intake would have any effect on protein deposition rate in broiler chickens. Energy intake was kept constant on a protein-free basis in an attempt to make energy and amino acid intakes independent of each other.

The hypotheses proposed were, that (1) for broiler chickens between 200 and 1,600 g body weight (BW), protein deposition rate would increase (linearly) with increasing amino acid intake, until energy intake would become limiting; (2) additional amino acid intake above this point would be deposited less efficiently; and (3) an increase in energy intake would increase lipid deposition rate, but would not, at limiting amino acid intakes, affect protein deposition rate.

MATERIALS AND METHODS

Two experiments of similar design were performed with male broiler chickens over two body weight (BW) ranges, 200 to 800 g in Experiment 1

(EXP1) and 800 to 1,600 g in Experiment 2 (EXP2). The 1-d-old chicks for EXP1 and EXP2 were hatched on the same day and were housed in the same house.

Birds and housing

One hundred forty-three 10-d-old male broiler chickens (Ross 208, Ross breeders, New Bridge, UK) were individually housed in floor pens (1.0 by 0.36 m) on wood shavings. Temperature was gradually decreased each d from 26 C at Day 10 to 19 C at Day 42, at which level it was then held. Lighting was 16 h of light and 8 h of darkness.

Experimental design

The birds were assigned to 143 floor pens; three blocks of 21 pens (EXP1) and four blocks of 20 pens (EXP2), according to a randomised block design. Each block consisted of one replicate for each of the 18 treatments and, in addition, three (EXP1) or two (EXP2) *ad libitum* fed control birds. These control birds were used to estimate the intrinsic maximum protein deposition rate of the birds. Within blocks, variation in initial BW was minimized by allocation of birds based on BW. For the first 10 d of age, all broilers were housed in groups of 20 per pen.

Both experiments had 18 controlled feeding treatments: two levels of protein-free digestible energy supply ($\text{energy}_{\text{pf}}$ at 1.7 and 2.1 times estimated maintenance) combined with nine levels of amino acid supply (3.3, 3.8, 4.3, 4.5, 4.8, 5.0, 5.3, 5.7 and 6.2 (EXP1) and 3.2, 3.6, 4.1, 4.3, 4.6, 4.8, 5.0, 5.4 and 5.9 (EXP2) g apparently digestible lysine per Mcal $\text{energy}_{\text{pf}}$). The calculated nutrient levels were all based on CVB (2000). The estimation of AME requirement for maintenance (108 kcal per kg metabolic BW ($\text{kg}^{0.75}$) per d) came from the literature (Zoons *et al.*, 1991). The low and high levels of $\text{energy}_{\text{pf}}$ supply corresponded with, respectively, about 70% and 83% of the mean $\text{energy}_{\text{pf}}$ intake of the *ad libitum* fed birds, in both experiments. On the basis of the composition of ideal protein proposed by Baker *et al.* (1993), all essential amino acids were supplied at levels of at least 115% of that of lysine. It was then assumed that the responses to amino acid intake in this trial reflected a response to lysine as the first limiting amino acid.

Proteins, carbohydrates and fats differ in their ATP yield per Mcal AME (see Gerrits *et al.*, 1996). Therefore, in order to standardize the intake of energy available for maintenance and growth among the different amino acid

intake levels as much as possible, energy intake was kept constant on a protein-free basis and the ratio between digestible carbohydrates and digestible fats was kept constant. The energy_{pf} supply was estimated from the intakes of apparently digestible crude fat (9.28 kcal/g) and apparently digestible carbohydrates (4.14 kcal/g) (CVB, 2000).

Table 1. Composition of the experimental feeds

Ingredients, g/kg	Energy feed	Protein feed	Nutrients, g/kg	Energy feed	Protein feed
Corn	379.0	407.0	<i>Calculated:</i>		
Peas	75.0	75.0	DE protein-free ²	2544	2060
Soybean meal (490 g/kg CP)	45.1	210.6	Lysine ³	7.5	12.8
Toasted soybeans	102.2	-	Methionine ³	4.5	8.0
Sunflower meal	23.2	40.0	Cysteine ³	1.5	2.7
Wheat middlings	68.9	59.4	Threonine ³	5.4	9.5
Tapioca (650 g/kg starch)	180.0	32.4	Tryptophan ³	1.3	2.5
Animal fat	65.7	62.3	Isoleucine ³	5.5	9.7
Caseinate (830 g/kg CP)	11.0	75.1	Valine ³	6.6	11.7
Monocalcium phosphate	9.2	3.4	Leucine ³	10.3	19.2
Limestone	13.6	16.3	Arginine ³	8.7	15.4
Sodium bicarbonate	5.0	1.9	Histidine ³	2.9	5.6
Sodium chloride	1.0	1.0	Phenylalanine ³	5.5	10.4
Premix ¹	2.1	2.1	Dry matter	886	889
Choline chloride 40 g/kg	10.0	3.7	Crude protein	145	245
L-Lysine-HCl	2.16	-	Crude fat	106	89
DL-methionine	2.48	3.97	Ash	56	52
L-arginine	1.21	3.46			
L-isoleucine	0.78	0.31	<i>Analysed:</i>		
L-valine	1.33	0.88	Lysine	8.8	14.1
L-threonine	1.77	1.18	Methionine	4.6	8.1
L-tryptophan	0.11	-	Cysteine	2.2	3.4
			Threonine	6.0	9.9
			Dry matter	878	877
			Crude protein	143	240
			Crude fat	103	84
			Ash	58	53

1 Contributed per kg of diet: 22 mg Cu as CuSO₄, 15 mg Fe as FeSO₄, 19 mg Zn as ZnSO₄, 71 mg Mn as MnO, 0.8 mg Co as CoSO₄, 0.6 mg I as KI, 0.20 mg Se as Na₂SeO₃, 12,000 IU vitamin A (retinyl acetate), 2,500 IU vitamin D₃, 1.12 mg vitamin B₁, 5.6 mg vitamin B₂, 38 mg nicotinic acid, 11 mg d-pantothenic acid, 3.0 mg vitamin B₆, 0.06 mg d-biotin, 1.0 mg folic acid, 0.012 mg vitamin B₁₂, 30 IU vitamin E (dl- α -tocopheryl acetate), 2.1 mg vitamin K₃, 70 mg salinomycin, 50 mg zinc-bacitracin;

2 Energy in digestible carbohydrates and fats (kcal/kg) (CVB, 2000);

3 Apparently faecal digestible for poultry (CVB, 2000).

From Day 0 to 10, all birds received a commercial starter diet (crumble) for *ad libitum* intake. Water was provided *ad libitum* throughout. The *ad libitum* fed birds in EXP1 and EXP2 had *ad libitum* access to the feed with the amino acid level of 4.6 g apparently digestible lysine per Mcal energy_{pf}. This diet was also given *ad libitum* to the birds of EXP2, from Day 10 until about 800 g BW. All restrictedly fed birds were fed twice per day.

Each of the experimental feeds was made by mixing the two basal feeds (the 'energy' and 'protein' feed; 3 mm pellet; Table 1) in different ratios. The two levels of energy_{pf} intake were achieved by having two levels of allowance of each of the mixtures.

Weighing procedures

In order to calculate individual daily feed allowances, all birds were weighed three times a week and each time, their gain was predicted on basis of linear extrapolation for the next 2- or 3-d period. As weights approached 800 (EXP1) or 1,600 g (EXP2), the broilers were weighed twice daily just before feeding. They were killed when their BW exceeded 770 or 1570 g in EXP1 and EXP2, respectively. In EXP2, three (out of four) birds per treatment group were dissected. The fourth bird was a spare one in case of mortality.

Dissection procedures, chemical analyses and calculations

Birds were weighed, killed with an injection of 0.2 ml T61 (containing per ml: 250 mg embutramide, 50 mg mebezoniumjodide, 5 mg tetracainehydrochloride) in the wing vein and stored at 2 C for a maximum period of 7 d. The birds were not bled. At dissections, each bird was weighed, plunged into water of 60 C for 1 min, de-feathered, dried and weighed again for calculation of the weight of feathers by difference. Afterwards, the oesophagus, trachea, proventriculus, gizzard, intestines, heart, liver, gall bladder, kidneys, lungs, spleen and Bursa of Fabricius were dissected from the body. These organs were defined as the 'organ fraction'. The remaining body, including the abdominal fat pad, formed the 'carcass fraction'. The gastrointestinal tract was stripped of its contents. The carcass and organ fractions were weighed, homogenized in a blender and frozen.

The carcass and organ fractions of each bird were analysed in duplicate for dry matter, lipid, ash and nitrogen, according to the standards ISO 1442 (1973), 1444 (1973), 936 (1978) and 937 (1978), respectively, from the

International Organisation for Standardisation (Geneva, Switzerland). On average, 98.6% (SE 0.35) of the dry matter was accounted for by the sum of lipid, ash and N x 6.25. The small residual would be expected to be largely glycogen. Diets were analysed for dry matter, lipid, ash and nitrogen, according to ISO 6496 (1983), 6492 (1985), 5984 (1978) and 5983 (1979), respectively, also from the International Organisation for Standardisation. Protein in these samples was calculated as N x 6.25.

The total deposition of protein or lipid in the three fractions, carcass, organs and feathers, was calculated for each bird as the difference between the amounts at the start and at slaughter. The amounts at the start were estimated from the initial BW of the bird and the mean body composition of the reference group. The reference group for EXP1 consisted of seven broilers slaughtered at Day 10. The reference group for EXP2 consisted of the *ad libitum* fed broilers in EXP1, slaughtered at 800 g BW. Deposition rates were calculated as total deposition divided by the length of the experimental period. The protein content of the feathers was taken as 43.8%, based on Hancock *et al.* (1995). The increase in protein content of the feathers with age (Hancock *et al.*, 1995) was considered to be negligible over the range used here. Daily lysine intake for each bird was calculated from the individual feed consumption and total lysine content of their experimental diet.

Analysis of data

To test for the existence of a diphasic linear response in the relation between amino acid intake and protein and lipid deposition rate, a diphasic linear model was compared with a monophasic linear model. The diphasic linear model was derived from Koops and Grossman (1993):

$$Y = a + b_1 * X - (b_1 - b_2) * 0.01 * \ln\left(1 + \exp\left(\frac{(X - c)}{0.01}\right)\right) \quad [1]$$

where: Y = protein or lipid deposition rate, a = intercept, b₁ = slope of the first linear phase, X = average daily lysine intake of individual bird, b₂ = slope of second linear phase, c = point of transition for the independent variable. In this model there is a sharp transition from the first to the second linear phase. The monophasic linear model was as follows:

$$Y = a + b * X \quad [2]$$

where: Y = protein or lipid deposition rate, a = intercept, b = slope and X = average daily total lysine intake of individual bird. The significance of the difference between models [1] and [2] was assessed by an F test. The

difference in sum of squares of the residuals between the two models, divided by the difference in df of the residuals, was tested against the residual MS of model [1].

As model [1] was never significantly better than model [2], a monophasic linear response of protein and lipid deposition rate to amino acid intake was assumed in further analyses. The effect of energy_{pf} and of amino acid intakes on protein and lipid deposition rates in the different body parts were analysed according to the following model:

$$Y = \mu + B_i + E_j + \beta_1 * (X_{jk} - X_{aver}) + \beta_{2j} * (X_{jk} - X_{aver}) \quad [3]$$

where: Y = rate of protein or lipid deposition, μ = average of Y at X = X_{aver}, B_i = fixed effect of block i, E_j = fixed effect of energy_{pf} intake level j, β_1 = effect of amino acid intake level (regression coefficient), β_{2j} = interaction between amino acid level and energy_{pf} intake level (difference in regression coefficients between the two energy_{pf} intake levels), X_{jk} = average daily lysine intake of bird k at energy_{pf} intake level j, X_{aver} = average experimental daily lysine intake (0.60 and 1.20 g/d in EXP1 and 2, respectively), i = 1...3 (EXP1) or 1...4 (EXP2), j = 1, 2 and k = 1...27.

The effect of BW range on the linear relation between amino acid intake and protein deposition rate was analysed according to the following model:

$$Y = \mu + BW_i + \beta_1 * (X_{ij} - X_{aver}) + \beta_{2i} * (X_{ij} - X_{aver}) \quad [4]$$

where: Y = rate of protein deposition, μ = average of Y at X = X_{aver}, BW_i = fixed effect of BW range i, β_1 = effect of amino acid intake level (regression coefficient), β_{2i} = interaction between amino acid level and BW range (difference in regression coefficients between the two BW ranges), X_{ij} = average daily lysine intake of bird j at BW range i, X_{aver} = average daily lysine intake in EXP1 and 2 (0.90 g/d), i = 1,2 and j = 1...54.

Non-linear (Levenberg-Marquardt algorithm; Moré, 1977) and linear (GLM; SPSS, 1999) regression procedures were used to analyse the data.

RESULTS

The data of one bird in EXP1 and of three birds in EXP2 were omitted from the analyses due to sickness. As an indication of performance of the restrictedly fed birds, the following data are given. Average BW at the start

of EXP1 (200 – 800 g BW) was 208 g. The time taken to grow from 200 to 800 g BW varied between 12.5 and 29.0 d so that growth rate varied between 46.4 and 20.3 g/d. Feed conversion ratio varied between 1.24 and 1.98 g/g.

At the start of EXP2 (800 to 1,600 g BW), birds were either 22 or 23 d old. They took between 10.0 and 22.5 d to grow to 1,600 g with growth rates varying between 77.5 and 35.4 g/d. Feed conversion ratio varied between 1.47 and 2.32 g/g. In both experiments, growth rates increased and feed conversion ratios decreased with increasing intakes of amino acids or energy or both.

Protein deposition rate

Effects of amino acid and energy_{pf} intake Data on protein deposition rates in the carcass are presented in Figure 1. In the carcass, but also in other body parts (Table 2), protein deposition rate increased with increasing amino acid intake, in both EXP1 and EXP2.

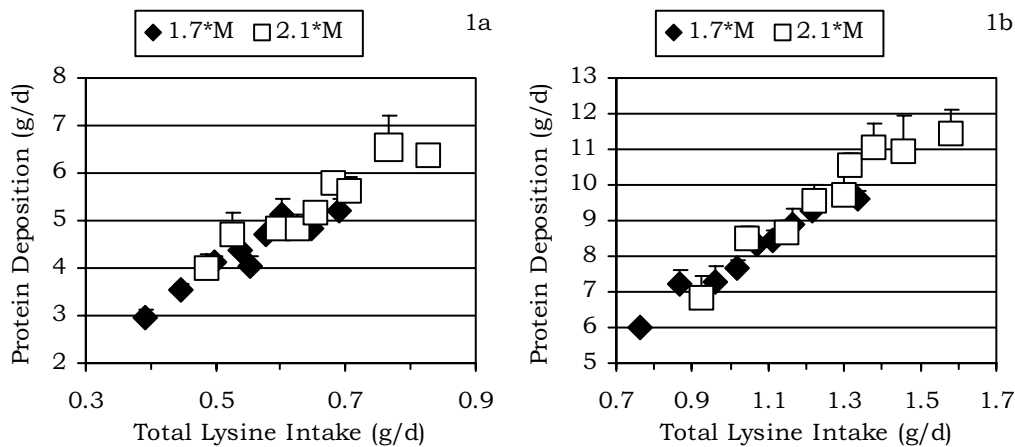


FIGURE 1. Protein deposition rate (g/d) in carcass of male broiler chicks, as a function of amino acid intake (total lysine as reference; g/d) and energy intake ($1.7 * M = 1.7$ times maintenance; $2.1 * M = 2.1$ times maintenance) at two BW ranges: 200 to 800 g (Figure 1a) and 800 to 1,600 g (Figure 1b). Error bars represent standard errors; $n =$ three replicates.

Parameter estimates for the diphasic model [1] are presented in Table 3. Model [1] did not provide a significantly better fit to the experimental data than did model [2], in either experiment (Table 3). There is thus no support

for the existence of a diphasic response in protein deposition rate, including a phase with a lower efficiency of protein deposition at high amino acid intake levels. At the high level of energy_{pf} intake in EXP1, model [1] did not converge.

Table 2. Effect of protein-free energy intake and amino acid intake (total lysine as a reference; g/d) on rate of protein deposition (g/d)

	Effect protein-free energy intake				Effect amino acid intake	Interaction ¹			
	Low _{2,3}	High _{2,3}	SED ⁴	P ⁵	P ⁶	Low ^{2,7}	High _{2,7}	SED ₄	P ⁸
<i>Experiment 1: 200 – 800 g BW</i>									
Total body	5.85	6.12	0.13	0.054	< 0.001	9.22	8.60	1.24	NS
Carcass	4.52	4.76	0.12	0.053	< 0.001	7.38	7.11	1.15	NS
Organs	0.62	0.64	0.024	NS	< 0.001	0.89	0.94	0.22	NS
Feathers	0.72	0.72	0.039	NS	< 0.001	0.95	0.55	0.36	NS
<i>Experiment 2: 800 – 1,600 g BW</i>									
Total body	11.5	11.8	0.29	NS	< 0.001	9.20	9.46	1.34	NS
Carcass	9.00	9.27	0.26	NS	< 0.001	6.31	6.97	1.20	NS
Organs	1.02	1.03	0.068	NS	< 0.001	1.31	1.04	0.31	NS
Feathers	1.55	1.50	0.082	NS	< 0.001	1.58	1.45	0.38	NS

1 Interaction exists if the effect of amino acid intake (expressed as the regression coefficient of the linear relation between amino acid intake and rate of protein deposition) differs for the low and high protein-free energy intake levels;

2 Low and high protein-free energy intake levels are 1.7 * M and 2.1 * M, respectively (M = energy for maintenance);

3 Values represent rates of protein deposition at the average total lysine intake (0.60 and 1.20 g/d in Experiment 1 and 2, respectively);

4 Standard error of difference;

5 Probability for test on effect of protein-free energy intake; NS: P > 0.10;

6 Probability for test if the regression coefficient of the linear relation between amino acid intake and rate of protein deposition, averaged over protein-free energy intake levels, equals zero;

7 Values represent regression coefficients of the linear relation between amino acid intake and rate of protein deposition;

8 Probability for test if a significant interaction exists between amino acid intake and protein-free energy intake; NS: P > 0.10.

Table 3. Parameter estimates (and asymptotic SE) for the diphasic linear relation¹ between amino acid intake (total lysine as a reference; g/d) and rate of protein deposition (g/d)

Protein deposition rate in:	Energy intake ²	a	b1	b2	c	R ² ³	P ⁴
<i>Experiment 1: 200 – 800 g BW</i>							
Total body	Low	- 0.27 (0.71)	10.9 (1.40)	3.95 (5.95)	0.61 (0.07)	0.85	NS
	High	- ⁵	-	-	-		-
Carcass	Low	- 0.43 (0.65)	8.80 (1.28)	2.85 (5.39)	0.61 (0.07)	0.81	NS
	High	-	-	-	-		-
<i>Experiment 2: 800 – 1,600 g BW</i>							
Total body	Low	0.02 (1.22)	9.76 (1.26)	7.57 (3.36)	1.15 (0.24)	0.89	NS
	High	-1.38 (2.21)	11.2 (1.93)	5.77 (3.89)	1.33 (0.15)	0.78	NS
Carcass	Low	0.89 (0.85)	6.84 (0.86)	3.73 (4.59)	1.21 (0.19)	0.85	NS
	High	-0.46 (1.96)	8.17 (1.70)	3.87 (4.47)	1.36 (0.20)	0.69	NS

1 $Y = a + b1 * X - (b1-b2) * 0.01 * \ln(1 + \exp((X-c) / 0.01))$; where Y = rate of protein deposition (g/d), a = intercept, b1 = slope of the first phase, b2 = slope of the second phase, c = point of transition for the independent variable and X = total lysine intake (g/d);

2 Low and high = 1.7*M and 2.1*M energy intake level, respectively (M = energy for maintenance);

3 Proportion of variance explained by the model;

4 Probability for test if the diphasic linear model fits the data equally well as a monophasic linear model; NS: P > 0.10;

5 The diphasic function did not converge for these data.

The effects of energy_{pf} and of amino acid intakes on protein deposition rate are given in Table 2. Energy_{pf} intake level generally had no significant ($P > 0.05$) effect on protein deposition rate, either in total body or in carcass, feathers or organs. Thus, at limiting amino acid intakes, protein deposition rates were usually not increased with additional energy_{pf} intake. For total body and carcass in EXP1, however, protein deposition rate, at limiting amino acid intakes, tended to be higher with the higher energy_{pf} allowance ($P < 0.10$). In EXP1 the difference in protein deposition rate in the total

body between the two levels of energy_{pf} allowance was on average 5% (Table 2). In EXP2, this difference was numerically similar to EXP1, but was not significant either. There was no interaction between the supplies of amino acids and energy_{pf} on protein deposition rate in total body, carcass, feathers or organs (Table 2). In other words, energy_{pf} intake did not significantly influence ($P > 0.05$) the increase of protein deposition rate (regression coefficient) with increasing amino acid intake.

The average daily lysine intake of the ad libitum fed birds was 0.75 g/d (SE 0.03) in EXP1 and 1.55 g/d (SE 0.04) in EXP2, with protein deposition rates in the carcass of 6.08 g/d (SE 0.13) in EXP1 and 11.14 g/d (SE 0.49) in EXP2. Protein deposition rates in total body were 7.85 g/d (SE 0.17) in EXP1 and 14.3 g/d (SE 0.61) in EXP2.

Effects of body weight range Data on the protein deposition rate in total body at the two BW ranges (EXP1 and EXP2 together) are presented in Figure 2.

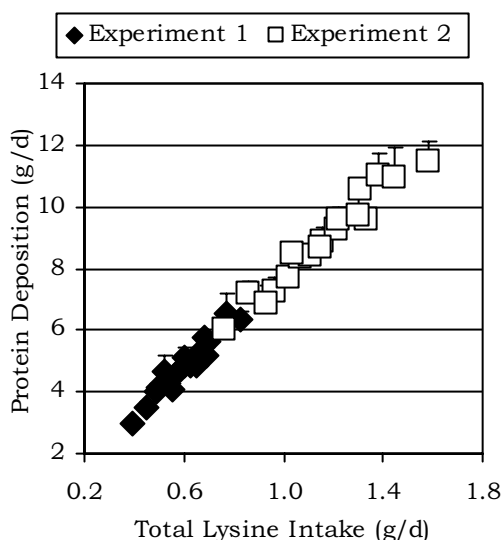


FIGURE 2. Protein deposition rate (g/d) in total body of male broiler chicks, as a function of amino acid intake (total lysine as reference; g/d), at two BW ranges (Experiment 1: 200 to 800 g; Experiment 2: 800 to 1,600 g). Error bars represent standard errors; $n =$ three replicates.

Data in Figure 2 shows that the relationship between protein deposition rate and amino acid intake is highly similar for the two BW ranges. In other words, at limiting amino acid intake, protein deposition rate seems independent of BW and to be determined solely by amino acid intake. This would imply that the regression coefficient (that is, the marginal efficiency of amino acid utilization for protein deposition) and the level of protein

deposition rate at a given amino acid intake level, in Figure 2 are not different between BW ranges. The marginal efficiency of protein deposition, defined as the increase in protein deposition rate with each additional unit of amino acid intake, was indeed not different ($P > 0.10$) between the two BW ranges (9.49 vs. 9.66 g protein per gram lysine for EXP1 and 2, respectively). The level of protein deposition rate at a given amino acid intake was also not different ($P > 0.10$) between the two BW ranges. Estimated protein deposition rates at 0.90 g lysine intake per d were 9.02 and 8.86 g/d for EXP1 and 2, respectively. If determined at amino acid intake levels much higher or lower than 0.90 g lysine per d, the level of protein deposition rate may be significantly different for the two BW ranges, given the numerical difference (NS) in marginal efficiency of protein deposition between BW ranges. Total efficiency of protein deposition, defined as protein deposition in total body divided by total lysine intake, was on average higher (t-test; $P < 0.01$) at the lower BW range than at the higher BW range (10.43 vs 9.88 g protein per gram lysine).

Lipid deposition rate

Results on the lipid deposition rates in carcass are summarized in Figure 3. Model [1] did not converge for lipid deposition rate. Thus, there was no evidence for the existence of a diphasic response in lipid deposition rate.

The effects of energy_{pf} and amino acid intake level on lipid deposition rate are given in Table 4. Lipid deposition rate was independent of amino acid intake. Level of lipid deposition rate was higher ($P < 0.001$) at the higher intake of energy_{pf}: 3.29 vs 1.87 g/d (EXP1) and 8.34 vs 5.09 g/d (EXP2) lipid deposition in carcass and organs for high and low energy_{pf} supply, respectively (Table 4). The effects of amino acid and energy_{p,f} supply on lipid deposition rate were similar for carcass and organs.

Lipid deposition rates in carcass of the *ad libitum* fed birds were 5.56 g/d (SE 0.18) and 11.85 g/d (SE 0.66) in EXP1 and 2, respectively. These rates of lipid deposition are much higher than the lipid deposition rates of the restrictedly fed birds with the higher energy_{pf} supply (Figure 3), in both EXP1 and EXP2. In total body, lipid deposition rates of the *ad libitum* fed birds were 5.88 g/d (SE 0.20) and 12.3 g/d (SE 0.65) in EXP1 and 2, respectively.

Table 4. Effect of protein-free energy intake and amino acid intake (total lysine as a reference; g/d) on rate of lipid deposition (g/d)

	Effect protein-free energy intake				Effect amino acid intake <i>P</i> ⁶	Interaction ¹			
	Low ^{2,3}	High ^{2,3}	SED ⁴	<i>P</i> ⁵		Low ^{2,7}	High ^{2,7}	SED ⁴	<i>P</i> ⁸
<i>Experiment 1: 200 – 800 g BW</i>									
Carcass + organs	1.87	3.29	0.17	< 0.001	NS	0.68	0.09	1.58	NS
Carcass	1.77	3.10	0.16	< 0.001	NS	0.72	-0.06	1.5	NS
Organs	0.10	0.18	0.014	< 0.001	NS	-0.04	0.15	0.13	NS
<i>Experiment 2: 800 – 1,600 g BW</i>									
Carcass + organs	5.09	8.34	0.57	< 0.001	NS	0.60	0.67	2.65	NS
Carcass	4.91	7.99	0.56	< 0.001	NS	0.72	0.69	2.59	NS
Organs	0.18	0.35	0.046	0.001	NS	-0.13	-0.02	0.21	NS

1 Interaction exists if the effect of amino acid intake (expressed as the regression coefficient of the linear relation between amino acid intake and rate of lipid deposition) differs for the low and high protein-free energy intake levels;

2 Low and high protein-free energy intake levels are 1.7 * M and 2.1 * M, respectively (M = energy for maintenance);

3 Values represent rates of lipid deposition at the average total lysine intake (0.60 and 1.20 g/d in Experiment 1 and 2, respectively);

4 Standard error of difference;

5 Probability for test on effect of protein-free energy intake;

6 Probability for test if the regression coefficient of the linear relation between amino acid intake and rate of lipid deposition, averaged over protein-free energy intake levels, equals zero; NS: *P* > 0.10;

7 Values represent regression coefficients of the linear relation between amino acid intake and rate of lipid deposition;

8 Probability for test if a significant interaction exists between amino acid intake and protein-free energy intake; NS: *P* > 0.10.

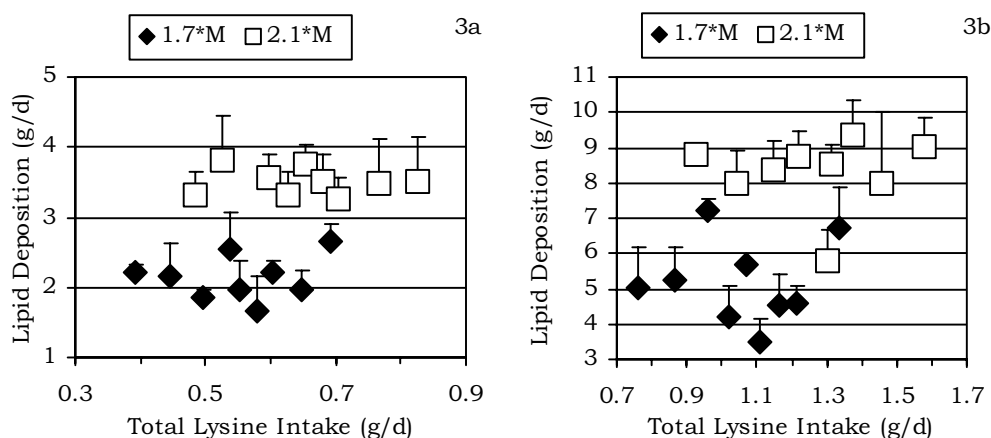


FIGURE 3. Lipid deposition rate (g/d) in carcass of male broiler chicks, as a function of amino acid intake (total lysine as reference; g/d) and energy intake (1.7 * M = 1.7 times maintenance; 2.1 * M = 2.1 times maintenance) at two BW ranges: 200 to 800 g (Figure 3a) and 800 to 1,600 g (Figure 3b). Error bars represent standard errors; n = three replicates.

DISCUSSION

Amino acid intake and protein deposition rate

Part of the hypothesis for this study was that protein deposition rate increases (linearly) with increasing amino acid intake, until energy intake becomes limiting for protein deposition rate. Additional amino acid intake above this point would be deposited less efficiently. Thus, a diphasic response in protein deposition rate was expected. In accordance with this hypothesis, diphasic response curves were found (Table 3), estimated by means of a diphasic model. However, the diphasic model did not fit the data significantly better than a monophasic model. Therefore, there was no statistical evidence that energy_{pf} intake had been limiting protein deposition rate at high amino acid intakes.

The power of the statistical test discriminating between the diphasic and monophasic model was low, which could have contributed to the failure to find a significant diphasic response. First, there were only two or three treatments (see Figure 1) above the estimated point of transition (Table 3).

As a consequence, the SE's of the estimated regression coefficients of the second phase were quite high (Table 3). Secondly, the difference in regression coefficients between the first and second phase of the diphasic model was relatively small. In our study, protein deposition rate did not reach a plateau, but continued to increase with increasing protein intake, although with a lower efficiency in the second phase than in the first phase. In contrast, trials of similar design have been reported (e.g. Bikker, 1994) where protein deposition rates reached a maximum (a plateau) at a certain protein intake. In such trials, the difference between the diphasic and monophasic model is more likely to be significant than in our study.

In the study of Bikker (1994), a plateau was found in protein deposition rate at high protein intakes, where energy intake became limiting. In his study, total AME intake, and not energy_{pf} intake, was kept constant among amino acid intake levels. In our study, with constant energy_{pf} intake, total AME intake increased with increasing amino acid intake. Consequently, protein deposition rate was less likely to become limited by energy intake.

Another possible reason exists for the absence of a significant diphasic response in protein deposition. Lipid deposition rate was not affected by increasing amino acid intake, at a constant energy_{pf} intake. Besides, daily maintenance requirement for energy was, most likely, not affected either by the increased amino acid intake, at constant energy_{pf} intake, because average body weight during the experiment was not different between treatments. Protein deposition rate, however, significantly increased with increasing amino acid intake, at constant energy_{pf} intake. These findings suggest that the energy required for additional protein deposition seems to have been supplied completely by (the partial oxidation of) the additional intake of amino acids. From this reasoning, it follows that energy_{pf} intake may have not been limiting protein deposition rate at all, not even at the highest amino acid intakes.

Even in perfectly balanced diets, some amino acid oxidation is inevitable (Millward *et al.*, 1990). However, in our study, amino acid oxidation was further increased due to the deliberate imbalance in dietary protein. All essential amino acids were supplied at levels of at least 115% of that of lysine. It follows that the oxidation of the amino acids was probably relatively high in our study.

Energy_{pf} intake and protein deposition rate

At limiting amino acid intake, additional energy_{pf} intake had generally no effect on protein deposition rate. This latter fact is accordance with several pig studies (e.g. Black et al., 1986; Kyriazakis and Emmans, 1992; Susenbeth et al., 1999) in which energy intake had no effect on protein deposition if protein intakes were limiting. In contrast, in a similar study with preruminant calves, additional energy_{pf} intake significantly increased protein deposition rates, although protein intake was limiting (Gerrits et al., 1996).

With young broiler chicks (EXP1), increasing the intake of energy_{pf}, at limiting amino acid intake, tended to increase protein deposition rate in total body and carcass. An explanation for this could be a 'protein-sparing' effect of additional energy_{pf} intake. Additional intake of carbohydrates may increase insulin levels in the blood, which may reduce amino acid oxidation (Hourani *et al.*, 1990), leaving more amino acids for protein deposition. On the other hand, the data as presented in Figure 1 do not really support the idea of a systematic effect of energy_{pf} intake on protein deposition rate in carcass. Taking together, at limiting amino acid intakes, additional energy_{pf} intake had generally no effect on protein deposition rate.

Birds fed ad libitum

Protein deposition rates of the restrictedly fed birds at the highest level of energy_{pf} and amino acid intake were in the same range as the protein deposition rates of the *ad libitum* fed birds. If it is assumed that the *ad libitum* fed birds grew according to their intrinsic maximum protein deposition rate, these facts suggest that the highest protein deposition rates of the restrictedly fed birds may have been close to their intrinsic maximum. Lipid deposition rates of the *ad libitum* fed birds were higher than the lipid deposition rates of restrictedly fed birds with similar protein deposition rates. This difference in lipid deposition rates is probably related to a difference in dietary protein to energy ratio.

Body weight and protein deposition rate

The partitioning of protein in the body among carcass, organs and feathers was different between BW ranges. For example, the proportion of total protein deposition that was deposited in organs decreased with increasing BW (results not presented). However, the marginal efficiency (=

the increase in protein deposition rate with each additional unit of amino acid intake) of protein deposition in total body was independent of BW. Thus, the difference in growth composition between BW ranges did not significantly modify the efficiency by which the dietary amino acids (above maintenance) were deposited in the body.

In contrast to the marginal efficiency, total efficiency (= total protein deposition divided by total lysine intake) of protein deposition (which includes maintenance losses) was significantly decreased with increasing BW. In EXP2, total efficiency of protein deposition in total body was 5% lower than in EXP1. Similarly, Susenbeth *et al.* (1999) found that, at a given intake of lysine as the first limiting nutrient, protein deposition rate in pigs decreased with increasing BW. At 77 kg BW, protein deposition rate was 5% lower than at 44 kg BW. The results of their and our study indicate that, if lysine intake is first limiting, maintenance requirement for lysine increases with BW. Absolute levels of maintenance requirement for lysine are low. In our study, the marginal efficiency of lysine for protein deposition was similar, or even lower, than the total efficiency. Based on this observation, estimated maintenance requirements for lysine are close to zero, or even negative. In the review of Susenbeth (1995), estimated maintenance requirements for lysine in pigs were not different from zero. In their and our study, maintenance requirements were estimated at zero protein retention. It is recognized, however, that using zero lysine retention as a parameter leads to much higher estimates of maintenance requirements (Emmert and Baker, 1997). This suggests that lysine needs for maintenance may be substantially higher than estimated by Susenbeth (1995) and in our study. Nevertheless, the estimated differences in maintenance requirement for lysine between BW ranges caused differences in protein deposition, at a given amino acid intake, that can not be ignored.

In our study, nearly all lysine intake levels in EXP2 were higher than in EXP1 (Figure 2). The effect of BW on total, and marginal, efficiency of amino acid utilization needs to be validated with data with more overlap in amino acid intake levels for the different BW ranges.

Conclusions

Protein deposition rate increased with additional amino acid intake. No evidence was found that energy_{pf} intake limited protein deposition rate at high amino acid intake. Extra intake of energy_{pf} increased lipid deposition rate, which was independent of amino acid intake. Where amino acid

intake was limiting, additional intake of energy_{pf} had generally no effect on protein deposition rate. The marginal efficiency of amino acid utilization for protein deposition did not depend on body weight. The facts are relevant to the modelling of the growth of broiler chickens.

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Chapter 2

**Fat-Free Body Composition in Male Broiler Chickens:
Effects of Feed Intake Level and Dietary
Protein to Energy Ratio**

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ABSTRACT The independence of fat-free body composition from nutrition is assumed in most models that simulate animal growth. This assumption has not been extensively investigated. In this study, firstly, the allometric relationships of water and ash with protein were investigated in growing broiler chickens. Secondly, it was tested if the amounts of water or ash at a given protein weight were affected by nutritional factors. Two experiments, each of a 2 x 9 factorial design, were conducted with male broiler chickens of two body weight ranges (200-800 g (EXP1) and 800-1600 g (EXP2)). The treatment factors were 2 levels of feed intake and 9 dietary protein to protein-free energy ratios (PE-ratio). Protein was balanced for amino acid content.

The allometric relationships of water and ash with protein were different for carcass and organs. The relationship between water and protein was not affected by nutrition, except for a 7% reduction in water weight at a fixed protein weight in the carcass in EXP1 at the lowest PE-ratio compared to the highest PE-ratio ($P < 0.001$). The relationship between ash and protein was strongly affected by nutrition. The lowest PE-ratio increased ash weight at a fixed protein weight in the carcass by up to 28%, compared to the highest PE-ratio ($P < 0.001$). It is concluded that, at least for modern meat-type animals, nutrition can significantly affect fat-free body composition at a certain fat-free body weight. The nutritional effects on fat-free body composition could be incorporated into models of the chemical body composition of growing animals.

INTRODUCTION

The prediction of the responses in growth rate and body composition at a given nutrient intake is a central problem in nutritional science. Models that simulate growth are a useful tool to address this problem as they can be used to integrate knowledge on nutrition and body growth and composition. The body tissues can be seen as being composed of protein, fat, water and ash (minerals), ignoring thereby a small amount of carbohydrates that will be present in the fed animal. In growth simulation models, protein and fat deposition are usually predicted from dietary energy and protein intakes. In most models, water and ash deposition are then associated with protein deposition only (Emmans, 1981; Black *et al.*, 1986; Moughan *et al.*, 1987). This implies that the relationships between water and protein, and between ash and protein, are both considered as independent of nutritional factors.

The relationships between water and protein, and ash and protein, in growing animals, have been described for several species, e.g. fattening pigs (Kotarbinska, 1969; cited by Moughan *et al.*, 1987), layer pullets (Kwakkel *et al.*, 1997) and turkeys (Emmans, 1989). However, the relationships were usually determined with animals growing according their potential growth curve under the conditions of *ad libitum* access to balanced diets. Literature is unclear whether, at restricted nutrient intakes, the relationships between water or ash and protein are similar to *ad libitum* conditions. Results for pigs (De Greef *et al.*, 1992) suggest that the relationship between water and protein is independent of feed intake level (FI-level) and of dietary protein to energy ratio (PE-ratio). On the proposed strong relationship between ash and protein, several studies report different results (Elsley *et al.*, 1964; Kyriazakis and Emmans, 1992). In some cases, the relationship between ash and protein seems independent of nutritional regime. A low PE-ratio, however, increased the ash to protein ratio in several pig studies (Zimmerman and Khajarern, 1973; Kyriazakis *et al.*, 1991). In the latter studies, the development of skeletal muscles might have been limited more by a low PE-ratio than the development of the skeleton. In that case, an effect of PE-ratio on the ash to protein ratio would be expected particularly in the carcass, more than in the organs. Information on possible nutritional effects on the relationships between water, ash and protein in body components is not

clear. It has implications for growth simulation models, as mentioned before, and for other subjects, such as meat quality (Berri, 2000).

The objectives of this study were (i) to investigate the allometric relationships of water and ash with protein in growing broiler chickens and, (ii) to analyse if these relationships are affected by nutritional factors. The second question was answered by testing if water and ash contents at a certain protein weight varied between dietary treatments. The hypothesis was that the amounts of water and ash at a certain protein weight, both in carcass and organs, are independent of both FI-level and PE-ratio. A second alternate hypothesis was that the amount of ash at a certain protein weight would be increased by a low PE-ratio, but only in the carcass and not in the organs.

MATERIALS AND METHODS

Experimental design

Detailed descriptions of the design of the experiments and of the dissection procedures have been presented elsewhere (Chapter 1). They are summarized here. Two experiments of similar design were performed with in total 126 individual floor pen housed male broiler chickens (Ross 208, Ross breeders, New Bridge, UK) in two body weight ranges: from 10-d-old (about 200 g) until 800 g (EXP1) and from 800 g until 1600 g (EXP2). Day-old chicks for both experiments were hatched on the same d and were housed in the same room. Each broiler chicken was assigned to either three (EXP1) or four (EXP2) blocks of 18 pens, according to a randomised block design. Each block consisted of one replicate for each of the 18 treatments. At d 10, seven additional birds were slaughtered and dissected as a reference for the body composition at the start of the experiment.

Both experiments consisted of 18 dietary treatments: two levels of feed intake combined with nine ratios of protein to energy. Protein was balanced for amino acid content. Feed intake levels were 1.7 and 2.1 times maintenance requirement for energy; energy was defined on a protein-free basis (see Chapter 1). Protein-free metabolisable energy was calculated from apparently digestible crude fat (38.83 kJ per g) and apparently digestible carbohydrates (17.32 kJ per g) (CVB, 2000). Metabolisable energy requirement for maintenance was estimated as 450 kJ per kg metabolic body weight ($\text{kg}^{0.75}$) (Zoons et al., 1991). Birds were fed twice per day. Protein-free energy intake

with the low and high feed intake level corresponded with, respectively, about 70% and 83% of the mean protein-free energy intake of ad libitum fed birds in the same experiments (Chapter 1; results not presented here).

Protein to energy ratios were 0.80, 0.91, 1.03, 1.08, 1.14, 1.20, 1.26, 1.37 and 1.48 (EXP1) and 0.76, 0.87, 0.98, 1.03, 1.09, 1.14, 1.19, 1.30 and 1.41 (EXP2) g apparently digestible lysine per MJ protein-free energy. The middle one of the nine ratios was assumed to be optimal for protein deposition, at least on ad libitum intake. Starting from ideal protein (Baker et al., 1993), all essential amino acids were supplied at levels of at least 115% of their requirements, relative to lysine. Therefore, the first-limiting amino acid for protein deposition was most likely always lysine. Broilers of EXP2 had ad libitum access to the diet with 1.09 g digestible lysine per MJ protein-free energy during the pre-treatment period (10-d-old until 800 g body weight). Each of the experimental feeds was made by mixing two basal feeds (an 'energy' and 'protein' feed; 3 mm pellet; see Table 1 in Chapter 1) in different ratios. An official Dutch committee on animal care and ethics approved the experimental protocol.

Data collection

Birds were killed with an injection of 0.2 mL T61 (see Chapter 1), not bled and stored at 2 °C for a maximum period of 7 d. At dissections, birds were de-feathered and the metabolic and digestive organs were dissected from the body. The gastrointestinal tract was stripped of its contents. The, so-called, 'organ' fraction consisted of oesophagus, trachea, proventriculus, gizzard, intestines, heart, liver, bile bladder, kidneys, lungs, spleen and Bursa of Fabricius, together. The remaining body, including abdominal fat pad, was defined as the 'carcass' fraction. In EXP2, 3 (out of 4) animals per treatment group were dissected. The fourth animal was a spare one in case of mortality. Carcass and organ fractions of each animal as well as the experimental diets were analysed in duplicate for dry matter, lipid, ash and nitrogen content, as described in Chapter 1. Protein content was calculated as 6.25 x N. Protein weight in the carcass was calculated by multiplying protein content in the carcass by the carcass weight. Protein weight in organs and weights of ash and water were calculated similarly.

Curve fitting and statistical analysis

The procedures for analysing the relationship between water and protein weight will be described below. For ash and protein, the same procedures were adopted.

Firstly, the relationships between water and protein weight in the different body parts (carcass, organs and carcass+organs) of growing broiler chickens were described. The data used were from broilers of about 200, 800 (EXP1) and 1600 (EXP2) g body weight. The data were not ideal for the purpose, as they were not equally distributed over the domain described. Curve fitting was performed using the allometric model of Huxley (1932), which describes the log-log linear relationship of two body components [$\ln(W) = \ln(a) + b \times \ln(P)$], in which: \ln = natural logarithm, W = water weight (kg), a = scale parameter, b = allometric slope and P = protein weight (kg).

Secondly, the effect of the dietary treatments on the amount of water at a certain protein weight was tested. This was tested separately for 800 g (EXP1) and 1600 g body weight (EXP2). Water weights as measured were recalculated to the mean protein weight at 800 g and at 1600 g (corrected water weights). Recalculation to a mean protein weight was necessary because birds were killed at a similar body weight and not at a similar protein weight. The details of the calculation are in the Appendix. The effect of dietary treatments (FI-level and PE-ratio) on the corrected water weights in the different body parts was analysed according to the following model:

$$W_{corr} = \mu + FI_i + \beta_1 \times \left(\left(\frac{P}{E} \right)_j - 1.12 \right) + \beta_{2i} \times \left(\left(\frac{P}{E} \right)_j - 1.12 \right) + \varepsilon_{ij} \quad [1]$$

where: W_{corr} = corrected water weight, μ = mean corrected water weight at $P/E = 1.12$, FI_i = fixed effect of FI-level i , β_1 = effect of P/E-ratio, β_{2i} = interaction between P/E-ratio and FI-level i , $(P/E)_j$ = PE-ratio of animal j , 1.12 = mean P/E-ratio (g digestible lysine per MJ protein-free energy), ε_{ij} = error, $i = 1, 2$ and $j = 1 \dots 27$.

All statistical analyses were performed with linear (GLM; SPSS, 1999) regression procedures. Significance was assigned at $P < 0.05$; tendencies were assigned at $0.05 < P < 0.10$.

RESULTS

Data of one bird in EXP1 and three birds in EXP2 were omitted from analysis due to sickness.

Allometric relationships

Relationships for water and ash weight with protein weight are presented in Figure 1. For each line in the graph, there are data of the three weight groups, corresponding with (from the left to the right side of the graph) the initial slaughter group, the animals of 800 g body weight, and the animals of 1600 g body weight. On the log-log scale, both of the relationships between water or ash and protein are essentially linear (Figure 1). Linearity on log-log scales is a necessary feature of an allometric relationship.

TABLE 1. Parameter estimates and goodness of fit criteria for the allometric relation between water and protein weight, or ash and protein weight in broiler chickens fed two levels of nine diets with different balanced protein to protein-free energy ratios¹

	Parameter estimates ¹					Goodness of fit	
	ln(a)	SE ²	b	SE	P ³	R ² ⁴	DW ⁵
<i>Water and Protein</i>							
Carcass	1.17	0.015	0.939	0.008	***	0.992	2.05
Organs	1.55	0.041	1.018	0.011	#	0.987	1.75
Carcass + organs	1.22	0.013	0.945	0.007	***	0.993	2.03
<i>Ash and Protein</i>							
Carcass	-1.87	0.048	0.969	0.024	NS	0.931	1.88
Organs	-2.42	0.069	1.075	0.018	***	0.967	1.92
Carcass + organs	-1.90	0.044	0.998	0.024	NS	0.937	1.84

1 $\ln(y) = \ln(a) + b * \ln(x)$; where ln = natural logarithm, y = water or ash weight (kg), a = scale parameter, b = allometric slope and x = protein weight (kg). N=115 for each allometric relationship.

2 SE = Standard error of estimate.

3 P = Probability for test if allometric slopes (=b) are different from unity; # $0.05 \leq P \leq 0.10$; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; NS, $P > 0.10$.

4 R² = Proportion of sum of squares explained by the model.

5 DW = Durbin Watson statistic: a value around 2 indicates no autocorrelation.

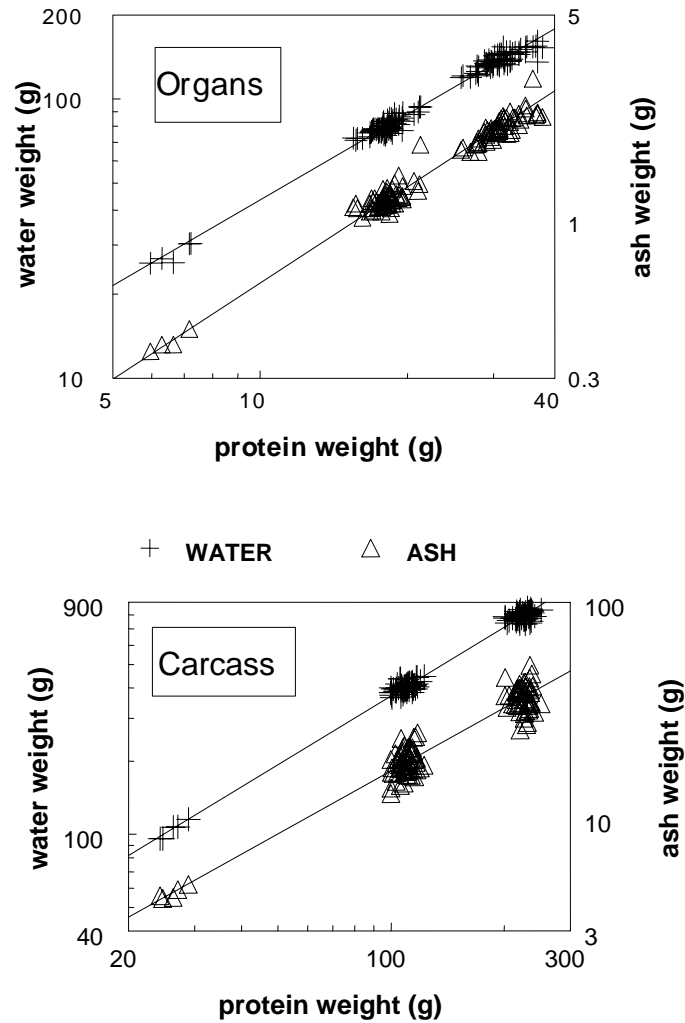


FIGURE 1. Relationships (log scale) for water and ash weight with protein weight in carcass and in organs of broiler chickens fed two levels of nine diets with different balanced protein to protein-free energy ratios. Values represent individual birds that were slaughtered at about 200, 800 or 1600 g body weight; $n=115$ for each relationship. All relationships were significant ($P < 0.001$). R^2 values for water and ash respectively, were 0.99 and 0.93 in carcass and 0.99 and 0.97 organs.

TABLE 2. Water and ash weight in different body parts of broiler chickens of 800 or 1600 g body weight with two feed intake levels (FI-level) of nine diets with different balanced protein to protein-free energy ratios (PE-ratio)¹

	FI-level				PE-ratio				Inter-action
	Low ²	High ²	SEM ³	P ⁴	Low ⁵	High ⁵	SE ⁶	P ⁷	P ⁸
-----water weight (g)-----									
Experiment 1 ⁹									
Carcass	406	403	15	NS	390	419	13	***	NS
Organs	79.7	80.4	2.7	NS	80.5	79.7	2.8	NS	NS
Carcass+organs	486	484	16	NS	471	499	14	***	NS
Experiment 2 ⁹									
Carcass	792	796	28	NS	789	799	28	NS	NS
Organs	139	138	5.6	NS	139	138	5.6	NS	NS
Carcass+organs	931	933	30	NS	927	936	30	NS	NS
-----ash weight (g)-----									
Experiment 1 ⁹									
Carcass	19.1	17.9	2.3	#	19.6	17.3	2.3	*	NS
Organs	1.22	1.20	.081	NS	1.24	1.17	.084	#	NS
Carcass+organs	20.3	19.1	2.3	#	20.9	18.4	2.3	*	NS
Experiment 2 ⁹									
Carcass	38.1	35.8	5.0	#	41.5	32.4	4.4	***	**
Organs	2.10	2.09	.097	NS	2.09	2.10	.099	NS	*
Carcass+organs	40.2	37.9	5.0	NS	43.7	34.3	4.4	***	**

1 Water and ash weights corrected to mean protein weight (18.1 and 31.1 g in organs and 110.6 and 225.2 g in carcass, in Exp.1 and 2, respectively).

2 Low and high FI-level are 1.7 x M and 2.1 x M, respectively (M = energy for maintenance).

3 SEM = Pooled standard error of the mean, n=54.

4 Probability for test on difference in water or ash weights between high and low FI-level; # 0.05 ≤ P ≤ 0.10; * P < 0.05; ** P < 0.01; *** P < 0.001; NS, P > 0.10.

5 Low and high PE-ratio represent least squares means for water or ash weight at lowest and highest PE-ratio in each experiment.

6 SE = Standard error of the estimate (linear regression between water or ash weight with PE-ratio), n=54.

7 Probability for test on (linear) relation of water or ash weight with PE-ratio; # 0.05 ≤ P ≤ 0.10; * P < 0.05; ** P < 0.01; *** P < 0.001; NS, P > 0.10.

8 Probability for test on interaction between FI-level and PE-ratio; # 0.05 ≤ P ≤ 0.10; * P < 0.05; ** P < 0.01; *** P < 0.001; NS, P > 0.10.

9 Experiment 1: birds slaughtered at about 800 g body weight; Experiment 2: birds slaughtered at about 1600 g body weight.

Parameter estimates for the allometric relationships of water and ash with protein are given in Table 1. Based on the goodness-of-fit criteria, the allometric relationship gives accurate descriptions of water or ash weight related to the total range of protein weights (Table 1).

For water versus protein in carcass+organs, the allometric slope was below unity (.945; Table 1). This means that, with increasing protein weights, the amount of water per unit of protein decreases continuously. In the carcass fraction, the allometric slope (.939) was also below unity. In the organ fraction, however, the allometric slope (1.018) was higher than unity. For ash in the carcass+organs, the allometric slope was very close to unity and not significantly different to it ($P > 0.10$). This implies that for this range of protein weights, the ash to protein ratio in the body did not change systematically with protein weights. This appears to be a consequence of an increasing ash weight per unit protein in the organs ($b=1.075$) combined with a decreasing (NS; $P > 0.10$) ash weight per unit protein in carcass ($b=.969$) as protein weight increases (Table 1).

Nutritional influences

Effects of feed intake level (FI-level) and dietary protein to energy ratio (PE-ratio) on water and ash weights at a given protein weight are given in Table 2. For all treatments, carcass weight accounted for about 86% of the weight of the carcass+organs fraction. Effects in the carcass+organs fraction thus mainly reflected effects found in the carcass fraction (Table 2).

Water weight at a given protein weight was unaffected by FI-level, both in carcass and organs, in EXP1 and EXP2. Water weight at a given protein weight was affected by PE-ratio, but only in the carcass in EXP1 ($P < 0.001$); at the lowest PE-ratio, water weight in the carcass was 390 g, which was about 7% lower compared to the highest PE-ratio (419 g; Table 2). In EXP2, this difference was only about 1% (NS).

Ash weight at a given protein weight was affected by FI-level, but only in the carcass ($P < 0.10$); at the low FI-level, ash weight at a given protein weight in carcass was about 6% higher than at the high FI-level, in both experiments (Table 2). Ash weight at a given protein weight was also affected by PE-ratio ($P < 0.05$; Table 2). In the carcass, a lower PE-ratio increased ash weight compared to a higher one; across the range the

difference was 13% in EXP1 and 28% in EXP2. In the organs, a low PE-ratio increased the ash weight by 6%, but only in EXP1 ($P < 0.10$). This latter effect was the only effect of the dietary treatments in the organs.

There were significant interactions between FI-level and PE-ratio with regard to ash weights in EXP2 (Table 2), both in carcass and organs ($P < 0.05$). In carcass, the difference in ash weight between low and high FI-levels was much larger at the lowest PE-ratio (44.9 vs. 37.0 g, respectively) than at the highest PE-ratio (29.7 vs 30.8 g, respectively). The background of the interaction in organs was that at the low FI-level, ash weight increased ($P < 0.01$) with increasing PE-ratio, whereas at the high FI-level, ash weight decreased ($P > 0.10$) with increasing PE-ratio.

DISCUSSION

The present study aimed to verify the theory that the water and ash contents in the body of growing animals only depend on the protein content in the body, irrespective the animals' diet. Water and ash weights in growing broiler chickens fed different diets in different quantities were compared. In an attempt to make comparisons at the same protein weight, broiler chickens were slaughtered at a fixed body weight. As a result, age at slaughter weight varied between dietary treatments, particularly between PE-ratios. Age at slaughter weight at the lowest and the highest PE-ratio were, respectively, 33.9 (se 1.57) and 24.8 (se 0.73) d in EXP1 and 42.1 (se 1.47) and 34.7 (se 0.49) d in EXP2. Differences in age at a body weight reveal variation in growth curve among the dietary treatments.

Even though all animals were slaughtered at a fixed body weight, protein weights differed systematically between dietary treatments. These differences in protein weight mainly reflected differences in fat weights between dietary treatments. In order to prevent that these differences in protein weight would interfere with the analysis for possible nutritional effects on the water to protein ratio or the ash to protein ratio, water and ash weights were re-calculated to a common (the mean) protein weight. This re-calculation was based on the relationship between water and protein, and ash and protein, within treatments. In this way, bias in the subsequent analyses for differences between treatments was prevented.

Water weights

The relationship between water and protein in the body of animals, at least at maturity, seems independent of genotype (Emmans, 1988; Wang *et al.*, 1999). Regarding the dependency of the relationship between water and protein on nutritional strategy, literature evidence is lacking. Kyriazakis and Emmans (1992) assumed that, under conditions that limit the growth of an animal, the water to protein ratio in a given component is not changed compared to that seen in normal growth. In other words, in the case of limited growth, deposition rates of water and protein are assumed to decrease in line with their inherent allometric relationship. For nutritional limitations of growth, this assumption is supported by the study of De Greef *et al.* (1992) with growing pigs, in which water weight at a certain protein weight seemed not affected by differences in FI-level and PE-ratio.

In our study, FI-level and PE-ratio did also not affect water weight at a certain protein weight, except in the carcass at 800 g body weight. The reduction in water weight in the carcass at 800 g body weight due to a low PE-ratio suggests that the assumption of Kyriazakis and Emmans (1992), mentioned above, may not be generally valid across genotypes or degrees of maturity of the animal. The anatomical background of the reduction in water weight in carcass could be a disproportional reduction of the growth of different tissues in the carcass, with different water to protein ratios, for example, a greater reduction in the growth of muscle protein compared to collagen protein (Ashgar *et al.*, 1986). Data of Gerrits *et al.* (1998) on the amino acid composition of pre-ruminant calves also suggested that the growth of muscle protein would be more inhibited by a low PE-ratio than would that of collagen protein. This proposed explanation, however, does not clarify why in older broilers, of 1600 g body weight, the effect of PE-ratio on water weight in carcass was not present.

Ash weights

Both in carcass and organs, a low FI-level and a low PE-ratio reduced the rate of protein deposition, by reducing the intake of lysine, the first limiting nutrient (Chapter 1). In the carcass, the rate of protein deposition was reduced more than the rate of ash deposition, resulting in an increased ash weight at a given protein weight. In organs, however, a low FI-level and a low PE-ratio caused, in most cases, a proportional reduction in rates of

protein and ash deposition, resulting in similar ash weights at the mean protein weight. A possible explanation for this difference between carcass and organs is that in the carcass, protein and ash are physically less related than in the organs. In the carcass fraction, ash is mainly in skeletal tissues and protein relatively more in non-skeletal tissues, whereas in the organ fraction, ash and protein are distributed more equally among the different tissues. Therefore, considerable effects in carcass on ash weight at a given protein weight most likely reflect changes in muscle to bone ratio. In apparent contrast with this idea on differences between carcass and organs, PE-ratio tended ($P < 0.10$) to affect ash weight at a given protein weight in organs also (at 800 g body weight; Table 2). It seems unlikely that the relationship between ash and protein in a given organ is affected by nutrition. Probably, the effect on ash weight at a given protein weight in the organ fraction reflects a disproportional reduction of the growth of different organs with different ash to protein ratios. For example, with decreasing PE-ratio, the proportion of kidneys in the organ fraction was significantly reduced in this study (data not shown).

Opposing views exist regarding nutritional effects on the relationship between ash and protein in the body of the animal. A low PE-ratio increased ash to protein ratio in piglets (Zimmerman and Khajjarern, 1973; Kyriazakis *et al.*, 1991) and in preruminant calves (Gerrits *et al.*, 1997), whereas a low FI-level induced similar effects in growing broiler breeders (Yu *et al.*, 1992; Katanbaf *et al.*, 1989). In contrast, the studies of Wilson (1954a; 1954b) with domestic fowl, of Elsley *et al.* (1964) with pigs and lambs and of Kwakkel (1994) with layer pullets, suggest that the relation between ash and protein, or skeleton and lean body, is independent of nutrition.

Discrepancies in results of these studies are probably related to differences between studies regarding animal characteristics, nutritional treatments and type of data analysis. For example, differences in genotype, length of the experimental period and age at the beginning of this period may explain the difference in results with the pigs. Zimmerman and Khajjarern (1973) and Kyriazakis *et al.* (1991) used relatively modern pig breeds, whereas Elsley *et al.* (1964) used data of pigs from before 1940. Muscle to bone ratio, or fat-free body composition, of well-fleshed modern breeds might respond more pronouncedly to dietary PE-ratio than might that with more traditional breeds. Besides, in the first two pig studies, pigs were

grown from about 5 kg to 12 or 23 kg body weight, whereas in the latter study, pigs were grown from 5 to about 83 kg body weight. With a relatively long experimental period, effects at young age might be partly compensated at a latter age. With regard to the nutritional treatments, whether an effect of FI-level or PE-ratio is found on ash weight at a certain protein weight may depend on which nutrient is limiting the growth. For example, if lysine intake is limiting protein deposition, as in our study, a low FI-level and a low PE-ratio both reduce the intake of the limiting nutrient, and may therefore cause similar effects on body composition. In contrast, in a case energy is the limiting nutrient for protein deposition, PE-ratio has less effect on the intake of the limiting nutrient than FI-level, and therefore, effects of PE-ratio and FI-level on body composition may be different. Finally, the kind of data analysis of Kwakkel (1994) differed from most other studies mentioned. Kwakkel (1994) analysed the development of body components during the experimental period, while most other studies analysed body composition only at the end of the experimental period. In the latter approach, it becomes more critical when exactly the experiment is finished, more than with the approach of Kwakkel (1994).

Kwakkel (1994) suggested that discrepancies between his results and those of Yu *et al.* (1992), regarding ash/protein (in)dependence, can be explained by whether feathers are or are not included in the carcass fraction. Our results, however, with defeathered carcasses also showed an effect of nutrition on ash weight at the mean protein weight. Thus, they do not sustain the suggestion of Kwakkel (1994).

We suggest that in studies on the relation between nutrition and fat-free body composition, effects of genotype (e.g. fleshiness of the animal) and nutrient composition (e.g. the limiting nutrient) should be considered explicitly.

Growth simulation models

The assumption in several growth models, that water and ash deposition are determined solely by protein deposition (Emmans, 1981; Black *et al.*, 1986; Moughan *et al.*, 1987) is not sustained by this study. Particularly with diets high or low in PE-ratio, considerable differences were found in the weights of water and ash at a given protein weight compared to diets with a more average nutrient composition. Simulating growth of chemical body components based on protein deposition only does not consider these

effects. For simulation of body weight gain based on protein deposition only, the effects on water weight are numerically of more importance than the effects on ash weight. The effect (6%) on water weight at 800 g body weight denotes an effect on body weight of 3 to 4%. For ash weight, even the large effect (27%) of PE-ratio on ash weight in carcass+organs at a body weight of 1600 g signifies an effect of less than 0.6% on body weight, and less than 1.2% on body weight gain between 800 and 1600 g. Thus, simulation of body weight gain or chemical body composition solely based on protein deposition might be accurate in case of *ad libitum* access to balanced diets, but can induce systematic errors in the simulations in case of low FI-levels or extreme PE-ratios. As the values of other variables in models, such as maintenance, heat loss, *ad libitum* food intake and physical body composition, are often related to body weight, simulation errors in body weight may have a wide impact on the accuracy of predictions by such models (Emmans and Kyriazakis, 1995).

Conclusions

The allometric relationships of water and ash with protein differed between carcass and organs. The relationship between water and protein was, in most cases, not affected by nutrition, whereas the relationship between ash and protein was strongly affected, particularly by PE-ratio. The extreme PE-ratios caused differences in ash weight at a given protein weight in the carcass by up to 28%. Results suggest that the muscle to bone ratio in carcass is dependent of nutrition. It is concluded that, at least for modern meat-type animals, nutritional strategy can have significant effects on fat-free body composition at a certain fat-free body weight, at least in the short term. The nutritional effects on fat-free body composition could be incorporated into models of the chemical body composition of growing animals.

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APPENDIX

Water weights as measured were corrected to the mean protein weight at 800 g (EXP1) and 1600 g body weight (EXP2). This was done by means of a linear model (equation [i]), assuming a linear relationship between water and protein weight for the small range of protein weights within EXP1 and EXP2. Corrected water weights in carcass and organs of the individual animals were calculated according to:

$$W_{corr} = W - c \times (P - \bar{P}) \quad [i]$$

where: W_{corr} = corrected water weight, W = measured individual water weight, c = correction factor, P = measured individual protein weight and \bar{P} = mean experimental protein weight. The values used for \bar{P} and c (four values each: for carcass and organs in EXP1 and EXP2) are presented below. Values for \bar{P} were 18.1 and 31.1 g in organs and 110.6 and 225.2 g in carcass, in EXP1 and EXP2, respectively. Values for parameter c , which is the regression coefficient between W and P at 800 (EXP1) or 1600 g body weight (EXP2), could not be estimated over the different dietary treatments, because values of P were systematically different for the dietary treatments. Therefore, values for parameter c in equation [2] were estimated based on the relation between W and P within dietary treatments, using the following regression equation:

$$(W_{ij} - \bar{W}_i) = c \times (P_{ij} - \bar{P}_i) \quad [ii]$$

where: c = correction factor, W_{ij} = water weight of animal j at dietary treatment i , \bar{W}_i = mean water weight at dietary treatment i , P_{ij} = protein weight of animal j at dietary treatment i and \bar{P}_i = mean protein weight at dietary treatment i , $i = 1..18$ and $j = 1,2,3$. For the correction of water weights, estimated values for c were 3.84 and 2.91 in organs and 1.21 and 1.21 in carcass, in EXP1 and EXP2, respectively. For the correction of ash weights, estimated values for c were 0.061 and 0.060 in organs and 0.116 and -0.141 in carcass, in EXP1 and EXP2, respectively. Corrected water weights (W_{corr}) in carcass+organs were calculated for the individual animal as the sum of W_{corr} in carcass and W_{corr} in organs. For the correction of ash weight, the same procedures were adopted as for the correction of water weight.

Chapter 3

Dietary Balanced Protein in Broiler Chickens: Effects of Early Life Protein Nutrition on Later Responses

Paper by: R. M. Eits, R. P. Kwakkel, M. W. A. Verstegen and C. G. Emmans, 2003. Br. Poult. Sci. 44:398-409. Reproduced with permission of Taylor and Francis.

ABSTRACT A study was conducted with modern broiler chicks to test the effects of early life protein nutrition and sex on responses in growth and body composition to dietary protein at later age. Effects on the incidence of metabolic disorders were also evaluated. From 11 to 26 d of age (EXP1), birds were given 8 diets varying in balanced protein to energy ratio (BPE ratio) between 0.575 and 1.100 g digestible lysine per MJ AMEn. Birds from two treatment groups in EXP1 (BPE ratio of 0.725 and 1.025 g/MJ, respectively) were subsequently used in a test from 26 to 41 d of age (EXP2). In EXP2, 8 diets were fed varying in BPE ratio between 0.500 and 1.025 g/MJ.

Responses in weight gain and feed conversion to BPE ratio in EXP2 changed considerably when BPE ratio in EXP1 was modified, irrespective of sex. Up to 10% improvement in both weight gain and feed conversion in EXP2 was observed if BPE ratio in EXP1 was 0.725 compared with 1.025 g/MJ. With males, however, the effect of treatment in EXP1 on weight gain in EXP2 was present only at high BPE ratios. For the relative gain of breast meat and abdominal fat, but not for carcass, the responses of male broilers to BPE ratio in EXP2 were altered by the BPE ratio in EXP1. With females, responses in composition of the gain to diet in EXP2 were independent of BPE ratio in EXP1. The incidence of metabolic disorders was low, irrespective of treatment in EXP1. The lower BPE ratio in EXP1 increased mortality in EXP2 from 0.8 to 3.6%.

Our findings show that broiler responses to dietary protein depend on previous protein nutrition and sex. Effects of early life protein nutrition on incidence of metabolic disorders were not observed. The results strongly suggest that protein levels in grower and finisher diets should not be optimised independently, but simultaneously.

INTRODUCTION

Determining which protein level is optimal in broiler diets, either for maximising broiler performance or profit, requires knowledge on how broiler performance and carcass quality are related to dietary protein level. Such knowledge can be obtained from experiments in which different doses of dietary protein are supplied over a given age period (for example, Clark *et al.*, 1982; Smith *et al.*, 1998). Where the data come from a later age period any effects of the protein level in the diet used previously are usually, but not always (Pesti and Fletcher, 1984), ignored. The question raised here is to what extent dietary history affects the responses to dietary protein levels.

The dietary history may be particularly relevant when low protein diets are fed during the early grower phase. Recent work on ascites in broiler chickens showed that such a feeding strategy may improve the resistance of broilers to metabolic disorders at later ages (Scheele *et al.*, 1999). Broilers given a low protein diet at early ages have been shown to compensate in gain during the re-alimentation period (Moran, 1979; Pesti and Fletcher, 1984). The capacity for growth compensation is probably higher for males than for females (Plavnik and Hurwitz, 1988) and may be less with fast growing strains than with slower growing strains (Cherry *et al.*, 1978). However, literature on compensatory responses with modern broiler breeds is scarce or even not available. Subsequent feed efficiency may be improved after an early protein restriction (Moran, 1979; Pesti and Fletcher, 1984). This is most likely due to a lower fat content in the gain (Gous *et al.*, 1992). In addition, the growth curve becomes more convex and thus, in theory, maintenance requirements will be reduced (Zubair and Leeson, 1996). Total protein intake to reach a certain body weight may be reduced in such a feeding strategy, as was found with turkeys by Auckland and Morris (1971). Obviously, such a protein sparing effect has concomitant advantages for production costs and environmental nitrogen pollution.

In broilers, the optimal nutrient levels during the re-alimentation period are not well-known (Zubair and Leeson, 1996). Model calculations (Plavnik and Hurwitz, 1989) as well as some experimental data (Auckland and Morris, 1971; Plavnik and Hurwitz, 1989) suggest that a protein level higher than those conventionally used may be needed.

It can be hypothesised that, if a low protein level is used in the early grower phase, the performance level in the later grower phase will be increased. In addition, it is likely that any nutritional carry-over effects would depend on the sex of the birds, and that modern fast and efficient growing breeds do have limited ability to show compensatory growth. It is also hypothesised that broiler chickens will require relatively high protein levels in the later grower phase to enable compensatory response to be seen.

The objective of the present study was to test for modern broilers if, during the later grower phase, the responses in growth and body composition to the ratio of balanced protein to energy in the diet, depend on the value of that ratio in the diet fed during the early grower phase. Balanced protein indicates that dietary protein was balanced for amino acid levels. The incidence of metabolic disorders was also measured to find out whether feeding strategy affects the incidence of such health problems.

MATERIALS AND METHODS

From 11 to 26 d of age (EXP1), birds were fed eight diets varying in balanced protein to energy ratio (BPE ratio; g digestible lysine per MJ AMEn). Birds from two treatment groups in EXP1 (BPE ratio of 0.725 and 1.025 g/MJ, respectively) were subsequently used during the succeeding test period (26 to 41 d of age; EXP2) where they were fed eight different BPE ratios. Any difference in response in EXP2 between the two groups with a different diet in EXP1 would indicate that early life protein nutrition affects broiler responses to dietary BPE ratio at later age. The other six treatment groups in EXP1, apart from the treatment groups with a BPE ratio of 0.725 and 1.025 g/MJ, respectively, were used to evaluate in EXP1 the degree of deficiency or excess in protein supply with the latter two groups.

Sexes were penned separately. In EXP1, a total of 1632 11-d-old Hybro-G broiler chickens (Hybro B.V., Boxmeer, the Netherlands), 816 of each sex, were allocated to a room with 96 floor pens with 17 birds per pen. For each treatment, there were 3 replicate pens per sex; 48 pens in total. For each of the BPE ratios 0.725 and 1.025 g/MJ there were an additional 12 replicate pens per sex giving an extra 48 pens in total. The high number of pens for these two BPE ratios was needed, because the birds in these pens were to be used in EXP2.

In EXP2, a total of 960 birds, 480 of each sex, were allocated to 96 pens with 10 birds per pen. The 960 birds for EXP2 were randomly selected from two treatment groups in EXP1 (BPE ratios of 0.725 and 1.025 g/MJ, respectively), 480 birds from each group. For each of the 16 treatment groups in EXP2 (8 BPE ratios with each of the 2 groups with a different BPE ratio in the preceding period (EXP1)), there were 3 replicate pens per sex.

Birds had free access to feed and water. Room temperature was gradually decreased from 34°C at 0 d of age to 25°C at 17 d. It was then decreased further to 20°C at 41 d. The chickens were vaccinated for Newcastle disease at 0 and 19 d of age and for Gumboro at 14 d of age. Lighting regime was 23 h light per day. Each pen was 1.00 m by 0.80 m and the floor was covered with wood shavings.

Dietary treatments

The calculated balanced protein to energy ratios (BPE ratios) tested in EXP1 were 0.575, 0.650, 0.725, 0.800, 0.875, 0.950, 1.025 and 1.100 g/MJ, expressed as g apparent faecal digestible lysine per MJ AMEn. In EXP2, the values of that ratio were 0.500, 0.575, 0.650, 0.725, 0.800, 0.875, 0.950 and 1.025 g/MJ. Digestible lysine was kept constant at 4.8% of the crude protein. Contents of all other essential amino acids, relative to lysine, were in accordance with the composition of ideal protein (as proposed by Baker and Han (1994)) or higher. Dietary energy was calculated as AMEn for broilers (CVB, 2000). Two basal diets were made (Table 1) that differed in protein and amino acid content. Each of the experimental diets was obtained by mixing the two basal diets in a certain ratio. The two basal diets were analysed for dry matter, lipid, ash, nitrogen, and total amounts of several minerals (Ca, P, Na, K and Cl), according to the standards ISO 6496 (1983), 6492 (1985), 5984 (1978) and 5983 (1979), respectively, from the International Organisation for Standardisation (Geneva, Switzerland). Crude protein in these samples was calculated as N x 6.25. In addition, total amino acid concentrations in the basal diets were analysed according to Llamas and Fontaine (1994). All nutrient analyses agreed well with the calculated values. From 0 to 11 d of age, all birds received a commercial starter diet (11.5 MJ AMEn-broilers, 210 g/kg crude protein, 10.5 g/kg digestible lysine).

Table 1. Composition of the basal diets (g/kg)

Ingredients	High protein diet	Low protein diet	Nutrients, calculated	High protein diet	Low protein diet
Wheat	388.4	333.7	AMEn (MJ/kg) ²	12.0	12.9
Corn gluten (600 g/kg CP)	78.7	-	Dry matter	891	891
Wheat bran	95.6	137.4	Crude protein	271	133
Soybean meal (490 g/kg CP)	199.2	57.8	Crude fat	102	136
Sunflower meal (340 g/kg CP)	38.8	10.0	Crude fibre	35	35
Potato protein	12.9	3.0	Ash	51	56
Peas	-	130.5	Calcium	7.7	7.8
Tapioca	-	149.4	Phosphorus	7.4	6.6
Fish meal	29.9	6.0	Phosphorus av. ²	3.9	3.9
Meat meal (570 g/kg CP)	51.8	14.9	Sodium	1.7	1.6
Animal fat	28.9	48.8	Potassium	9.1	7.1
Soya oil	47.8	70.7	Chloride	2.1	2.1
Sodium bicarbonate	2.8	2.4			
Limestone	7.6	10.5	BPE ratio ³	1.10	0.50
Monocalcium phosphate	3.3	11.5	Lysine ⁴	13.2	6.4
Sodium chloride	-	1.7	Methionine ⁴	6.4	3.0
DL-Methionine	1.9	1.3	Met + Cys ⁴	9.9	4.8
Lysine	2.7	1.1	Threonine ⁴	9.3	4.5
Threonine	0.6	0.7	Tryptophan ⁴	2.5	1.2
Biofeed mix (60 g/kg)	4.0	4.0	Arginine ⁴	13.9	7.6
Vitamin / mineral premix ¹	4.0	4.0	Valine ⁴	10.9	4.9
Choline (500 g/kg)	0.9	1.0	Isoleucine ⁴	9.8	4.3
			Histidine ⁴	5.4	2.6
			Leucine ⁴	21.1	7.7

1 Contributed (in mg per kg of diet): 22 Cu as CuSO₄, 15 Fe as FeSO₄, 19 Zn as ZnSO₄, 71 Mn as MnO, 0.8 Co as CoSO₄; 0.6 I as KI, 0.20 Se as Na₂SeO₃, 4.13 retinyl acetate, 0.063 cholecalciferol, 1.2 thiamine, 5.6 riboflavine, 38 nicotinic acid, 11 d-pantothenic acid, 3.0 pyridoxine, 0.061 d-biotin, 1.0 folic acid, 0.012 mg cyanocobalamin, 30 dl- α -tocopheryl acetate, 2.1 menadione, 70 salinomycin, 10 avilamycin.

2 For broiler chickens (CVB, 2000).

3 Balanced protein to energy ratio (g digestible lysine per MJ AMEn).

4 Apparent faecal digestible for broiler chickens (CVB, 2000).

Dissection procedures and calculations

Broiler weights at 11, 26 and 41 d, and feed intakes during the 11 to 26 and 26 to 41 d periods were recorded per pen. Feed conversion ratios were calculated for the respective periods. At 26 and 41 d of age, two birds per

pen were randomly selected for dissection and killed by CO₂-anaesthetic, according to approved ethical standards. For BPE ratios 0.725 and 1.025 g/MJ at d 26, 6 pens per treatment were randomly selected for dissection, instead of 3 pens per treatment in all other cases. Body weights and weights of carcass, breast meat (without skin) and abdominal fat pad were measured. The abdominal fat pad included the fat surrounding the gizzard and was used as an indicator of total body fat. Carcass weight was defined as the weight of the whole body, without skin, skin fat, feathers, head and organs (digestive organs, heart and lungs). In addition, the distal parts of the wings and legs were also removed, as is common practice in commercial slaughterhouses. In order to have the gastro-intestinal tract of the birds almost empty at dissection the lights were switched off during the 8 h before slaughter and dissection.

Four variables were used to measure the incidence or degree of (sub-) clinical metabolic disorders. They were measured for each dissected animal at 41 d. The variables measured were arterial pressure index (API) and scores for heart fibrinogen, pericardial moisture and liver cirrhosis. API is the ratio of the weight of the right heart ventricle wall to that of both ventricles and the septum (Huchzermeyer and De Ruyck, 1986). The other three measures were scored by the same experienced person for all birds on a 4-point scale, where 0 = normal and 3 = very abnormal.

Breast meat yield was calculated by dividing breast meat weight by carcass weight. Yields of carcass and abdominal fat pad were calculated by dividing the weights of carcass and abdominal fat pad by body weight. Estimates of body composition per pen were calculated as the average of the yields of the dissected animals in that pen. Similarly, the measures of metabolic disorders were averaged per pen before data analysis. Feed intake data were corrected for the estimated intakes of the animals that died, on the basis of their body weights.

Statistical analysis

Responses to dietary BPE ratio are described by the following exponential model (Lehmann *et al.*, 1996):

$$Y = a + (k - a) * (1 - e^{-c*(X-0.575)}) \quad [1]$$

where: Y = broiler performance variable; a = intercept (that is, performance on the diet with BPE ratio = 0.575 g/MJ); k = maximum response (asymptote); c = curvature steepness; 0.575 = lowest BPE ratio

that EXP1 and EXP2 had in common; X = IPE ratio (g digestible lysine per MJ AMEn). When the exponential model did not converge, a simple linear model ($Y = a + k * (X-0.575)$; where k = slope and Y , a and X have the same meaning as in model [1]) was applied.

Carry-over effects of the BPE ratio in EXP1 on the responses to BPE ratio in EXP2 were tested. This was done by testing whether a separate dose-response curve for the two groups with different BPE ratio in EXP1 (model [2]) would give a significantly better fit than only one dose-response curve (model [1]) for these two groups together. Model [2] was:

$$Y = s_1 * [a_1 + (k_1 - a_1) * (1 - e^{-c_1 * (X-0.575)})] + s_2 * [a_2 + (k_2 - a_2) * (1 - e^{-c_2 * (X-0.575)})] \quad [2]$$

where: s_1 and s_2 are dummy variables. For the higher BPE ratio in EXP1 s_1 is 1 and s_2 is 0. For the lower BPE ratio in EXP1 s_1 is 0 and s_2 is 1. The parameters a_1 and a_2 , k_1 and k_2 , and c_1 and c_2 correspond to the groups fed the diets with the high and low BPE ratio in EXP1, respectively. These variables have similar meanings as in model [1]. The significance of the difference between model [1] and model [2] was assessed by an F-test. The difference in the sum of squares of the residuals of model [1] and [2], divided by the difference in degrees of freedom of the residuals, was tested against the residual mean square of model [2].

Effects of sex, BPE ratio in EXP1 and BPE ratio in EXP2 on health parameters and mortality were evaluated by analysis of variance. The discontinuous score data on heart fibrinogen, pericardial moisture and liver cirrhosis were first translated to a parameter with a normal distribution. Each score value was replaced by a x-value from the standard normal distribution, depending on the total percentage of animals with that score. This was based on the assumption that the scored characteristics have an underlying continuity with a threshold, which imposes a discontinuity on the visible expression of the character which was scored (Falconer, 1960). Arcsine transformation was applied to the mortality data to normalise the data. Pen was the experimental unit for all analyses except for the regressions. Regression models were fitted to the treatment averages of the experimental data. Non-linear (Levenberg-Marquardt algorithm; Moré, 1977) and linear (GLM; SPSS, 1999) regression procedures were used to analyse the data.

RESULTS

Dose-related responses in EXP1

Broilers in EXP1 (11 to 26 d) clearly responded to increasing dietary balanced protein to energy ratios (Figure 1). All performance characteristics, except breast meat yield, showed a dose-response effect from the lowest up to the highest BPE ratio in the test. Breast meat yield showed little or no improvement when BPE ratio was increased above 0.725 g/MJ. Parameter estimates for the exponential model fitting the responses in broiler performance in EXP1 are given in Table 2.

Two treatment groups from EXP1 (with BPE ratios of 0.725 and 1.025 g/MJ, respectively) were used to test the carry-over effect of early life protein nutrition on broiler responses in EXP2. Broilers fed the lower instead of the higher BPE ratio (0.725 instead of 1.025 g/MJ) had lower body weights and carcass yields, and contained relatively more abdominal fat at the beginning of EXP2 (Table 3). With males, the effect of BPE ratio on the relative abdominal fat pad weight was not significant ($P > 0.10$). Breast meat yield was similar for the two treatment groups. In both sexes, feed conversion ratio was 12 to 15 points higher with the lower instead of the higher BPE ratio in EXP1 (Table 3). Based on the estimated asymptotes in performance in EXP1 (Table 2), final body weight, feed conversion ratio, and carcass and breast meat yield at the higher BPE ratio in EXP1 (1.025 g/MJ) deviated less than 5% from the estimated maximum or minimum. Relative weight of abdominal fat pad in these broilers, however, was 15% (males) to 26% (females) higher than the estimated minimum.

Dose-related responses in EXP2

As in EXP1, the broilers in EXP2 responded markedly to increasing BPE ratios in the diet (Figures 2 and 3). Male broilers that had been given the high protein diet in EXP1 did not show any response in breast meat yield with increasing BPE ratio in EXP2 and did not increase body weight gain at BPE ratios above 0.575 g/MJ (Figure 2).

Parameter estimates for the exponential responses (model [2] only) in EXP2 and the related R^2 values, are given in Table 4. The probability values of the dose-related responses in EXP2 depending on the diet that was fed in EXP1 are also given.

Table 2. Parameter estimates (SE between brackets) for the exponential¹ responses of broiler chickens to dietary BPE-ratio² in EXP1 (11-26 d of age)

	Males				Females			
	a	k	c	R ²	a	k	c	R ²
Final body weight (BW; g)	1151 (11)	1372 (53)	2.62 (1.2)	.97	1018 (14)	1190 (61)	2.69 (1.8)	.93
Body weight gain (g/d)	55.8 (.60)	69.0 (2.0)	3.20 (.97)	.98	48.4 (.77)	60.6 (4.2)	2.45 (1.5)	.95
Feed conversion	1.757 (.009)	1.357 (.015)	4.76 (.47)	.99	1.725 (.007)	1.416 (.011)	4.80 (.47)	.99
Carcass yield (g/g BW)	.535 ³ (.004)	.042 ³ (.011)	-	.70	.513 (.005)	.561 (.009)	4.61 (2.2)	.91
Breast meat yield (g/g carcass)	.262 (.008)	.284 (.005)	12.0 (13)	.53	.264 (.007)	.291 (.008)	5.90 (5.7)	.66
Abdominal fat pad (g/g BW)	.043 (.003)	.024 (.004)	5.4 (3.3)	.84	.040 (.002)	.019 (.009)	2.77 (2.1)	.90

1 $Y = a + (k-a) * (1 - e^{-c * (x - 0.575)})$ [model 1; see text]; where: y = broiler performance variable; a = intercept (i.e. performance on the diet with BPE-ratio=0.575 g/MJ); k = maximum response (asymptote); c = curvature steepness; 0.575 = lowest BPE-ratio that was tested in both EXP1 and EXP2 (26-41 d of age); x = IPE ratio (g digestible lysine per MJ AMEn).

2 Balanced protein to energy ratio (g digestible lysine per MJ AMEn).

3 If the iterative algorithm that was used to estimate the parameters of the exponential model failed to converge, a simple linear model was used instead [$y = a + k * (x - 0.575)$]; where k = slope and y, a and x have the same meaning as in the exponential model.

Table 3. Performance of broiler chickens at 26 d of age fed a low or high dietary BPE-ratio ¹ in EXP1 (11-26 d of age)

	Males				Females			
	High BPE-ratio	Low BPE-ratio	SEM ²	P ³	High BPE-ratio	Low BPE-ratio	SEM ²	P ³
Final body weight (BW; g)	1296	1238	6.1	.001	1148	1087	7.0	.006
Feed conversion (11-26 d)	1.40	1.55	.005	< .001	1.44	1.56	.005	< .001
Carcass (g)	729	664	8.9	.001	630	578	9.8	.005
Carcass yield (g/g BW)	.562	.534	.005	.004	.552	.539	.004	.075
Breast meat (g)	210	184	4.3	.002	183	161	5.4	.016
Breast meat yield (g/g carcass)	.287	.278	.005	NS	.291	0.278	.005	NS
Abdominal fat pad (g)	35.6	40.4	2.7	NS	27.3	32.5	1.8	.078
Abdominal fat pad (g/g BW)	.028	.033	.002	NS	.024	.030	.002	.026

¹ Balanced protein to energy ratio (g digestible lysine per MJ AMEn-broilers).

² Pooled standard error of the mean; n=6 per treatment, except for BW and feed conversion ratio: n=15 per treatment.

³ P value of difference between high and low BPE-ratio; NS: P > 0.10.

Table 4. Parameter estimates (SE between brackets) for the exponential¹ responses of broiler chickens to dietary BPE-ratio² in EXP2 (26-41 d of age)

Sex	Response parameter	Low BPE-ratio in EXP1 (11-26 d of age)			High BPE-ratio in EXP1 (11-26 d of age)			R ² 3	R ² 4	P 5
		a	k	c	a	k	c			
Male	Final body weight (g)	2139 (29)	2388 (126)	3.62 (3.1)	2262 (53)	2267 (22)	45.2 (154)	.75	.64	***
	Body weight gain (BWG; g/d)	62.0 (1.4)	77.3 (5.3)	3.86 (2.3)	64.8 (2.4)	66.6 (1.1)	21.3 (20)	.85	.67	NS
	Feed conversion	2.229 (.017)	1.772 (.073)	3.67 (1.0)	2.269 (.018)	2.020 (.042)	5.18 (1.7)	.97	.82	**
	Carcass gain (g/g BWG)	.613 (.011)	.642 (.0068)	14.7 (6.4)	.585 ⁶ (.007)	.125 ⁶ (.028)	- ⁶	.85	.64	NS
	Breast meat gain (g/g carcass gain)	.285 (.012)	.322 (.028)	5.21 (7.6)	.293 ⁶ (.011)	.0011 ⁶ (.044)	- ⁶	.33	.12 ⁷	***
	Abdominal fat pad gain (g/g BWG)	.049 (.0038)	.0311 (.0036)	9.51 (4.0)	.0373 (.0030)	.0150 (.010)	4.19 (3.4)	.88	.64	***

Female	Final body weight (g)	1870 (15)	1948 (22)	6.89 (4.0)	1892 (13)	2197 (744)	0.96 (2.8)	.84	***
	Body weight gain (BWG; g/d)	53.4 (-.46)	60.1 (1.2)	4.77 (1.7)	51.7 ⁶ (-.43)	14.2 ⁶ (1.7)	- ⁶	.94	*
	Feed conversion	2.183 (.030)	2.030 (.027)	9.75 (3.6)	2.339 (.024)	2.103 (.063)	4.83 (2.4)	.93	***
	Carcass gain (g/g BWG)	.577 (.0091)	.636 (.019)	5.54 (3.5)	.595 (.0092)	.651 (.018)	5.64 (3.6)	.84	NS
	Breast meat yield (g/g carcass gain)	.314 ⁶ (.008)	.0885 ⁶ (.031)	- ⁶	.312 (-.0080)	.350 (.055)	2.83 (6.22)	.56	NS
	Abdominal fat pad gain (g/g BWG)	.0603 (.0051)	.0401 (.0046)	9.75 (4.6)	.0565 (.0040)	.0387 (.012)	4.49 (5.5)	.79	NS

1 $Y = a + (k-a) * (1 - e^{-c*(x-0.575)})$; with separate parameter estimates for the groups with a low and a high BPE-ratio in EXP1, respectively (see model [2] in text); where: y = broiler performance variable; a = intercept (i.e. performance on the diet with BPE-ratio=0.575 g/MJ); k = maximum response (asymptote); c = curvature steepness; 0.575 = lowest BPE-ratio that was tested in both EXP1 and EXP2; x = IPE ratio (g digestible lysine per MJ AMEn).

2 Balanced protein to energy ratio (g digestible lysine per MJ AMEn).

3 R^2 values in case of two separate dose-response curves for the two groups with different diets in EXP1.

4 R^2 values in case of one exponential dose-response curve for the two groups with different diets in EXP1 together.

5 Level of significance for test if response curves are different for the two groups with different diets in EXP1; NS: $P > 0.10$; * $0.10 > P > 0.05$; ** $0.05 > P > 0.01$; *** $P < 0.01$.

6 If the iterative algorithm that was used to estimate the parameters of the exponential model failed to converge, then a simple linear model was used instead [$y = a + k * (x-0.575)$]; where k = slope and y, a and x have the same meaning as in the exponential model.

7 Using a linear model [$y = a + k*(x-0.575)$]; where k = slope and y, a and x have the same meaning as in the exponential model.

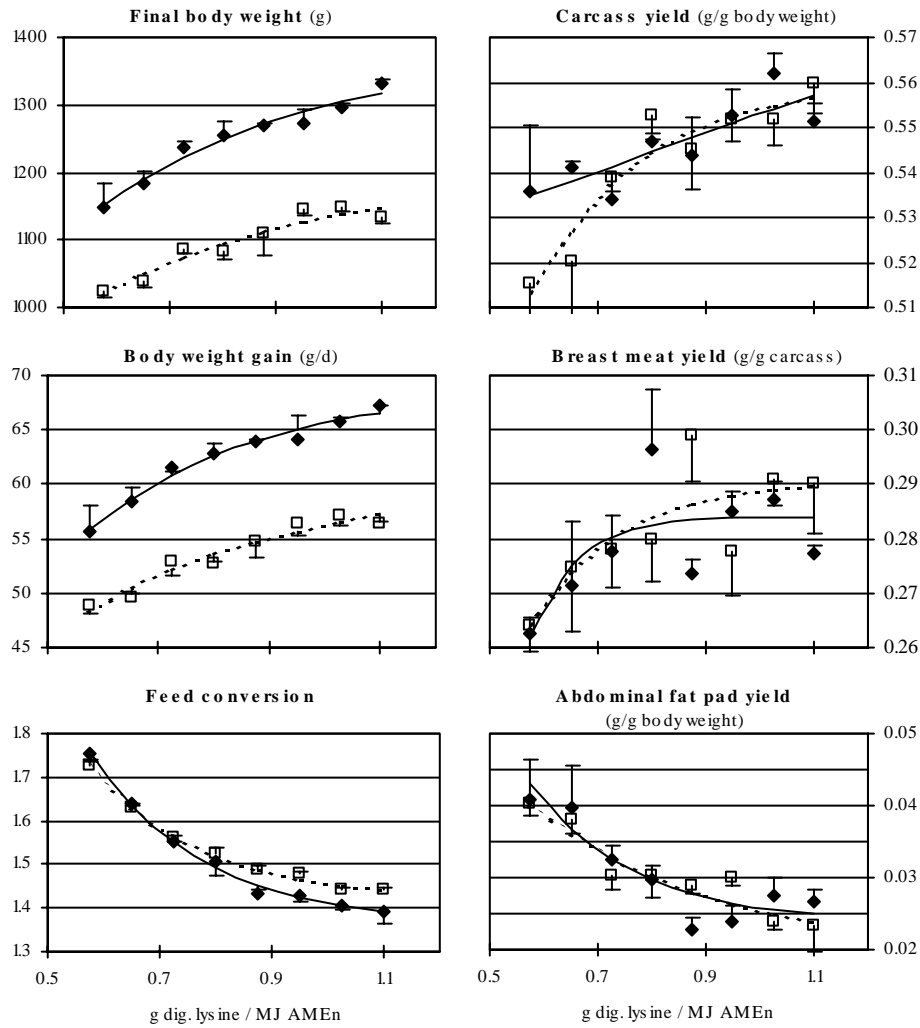


Figure 1. Dose-related responses of broiler chickens to dietary balanced protein to energy ratio (BPE-ratio) in EXP1 (11-26 d of age). Females (---, □) and males (—, ♦). Values are averages with SE indicated by the error bars ($n=3$, except for BPE-ratios 0.725 and 1.025 g/MJ, where $n=15$ for body weight, weight gain and feed conversion ratio, and $n=6$ for yields of carcass, breast meat and fat).

Despite lower body weights at the start of EXP2, broilers fed the low protein diet in EXP1 tended to have higher body weight gains in EXP2 than those fed the high protein diet in EXP1 (Figures 2 and 3). This difference in body weight gain was only significant for the females (Table 4). For the males, differences in body weight gain were present only at high BPE ratios in EXP2 (Figure 2). Provided that BPE ratios in EXP2 were high, male broilers fed the low protein diet in EXP1 compensated, at least to some extent, for their lower initial body weight (see final body weights in Figure 2). Female broilers fed a low protein diet in EXP1, however, did not fully compensate in EXP2 for their lower initial body weight (Figure 3).

Composition of the gain in females in EXP2 was unaffected by the diet in EXP1 (Figure 3, Table 4). With males, however, the proportion of abdominal fat in the gain and the content of breast meat in the carcass gain were affected by diet in EXP1. Males given a low instead of a high protein diet in EXP1, contained relatively more abdominal fat in the gain in EXP2 and, in spite of that, also utilised their feed in EXP2 more efficiently (Figure 2). This latter effect of diet in EXP1 on feed efficiency in EXP2 was also found in females (Figure 3).

Health status

Mortality averaged 0.6% in EXP1 and 2.4% in EXP2. Mortality was affected by sex ($P < 0.05$) with males contributing 96% and 76% in EXP1 and EXP2. Mortality in EXP2 was also affected by the diet that was fed in EXP1 ($P < 0.05$). Feeding the low instead of the high protein diet in EXP1 increased mortality in EXP2 from 0.8% (SE 0.40) to 3.6% (SE 0.77). Interactions of diet with sex, or between diet in EXP1 and EXP2 were not significant ($P > 0.05$).

Most of the metabolic variables were not affected by the experimental treatments. Liver cirrhosis was found in only one bird. The arterial pressure index was on average 0.192 (SD 0.018). The average heart fibrinogen score was low at 0.048 (SD 0.17). The average pericardial moisture score was higher at 0.44 (SD 0.39). Treatment did not affect arterial pressure index or heart fibrinogen score ($P > 0.05$). The pericardial moisture score, however, was significantly affected both by the diet in EXP2 ($P < 0.05$) and by sex ($P < 0.01$) (higher in males), but not by the diet that was fed in EXP1 ($P > 0.05$). Interactions between treatment factors were not significant ($P > 0.05$).

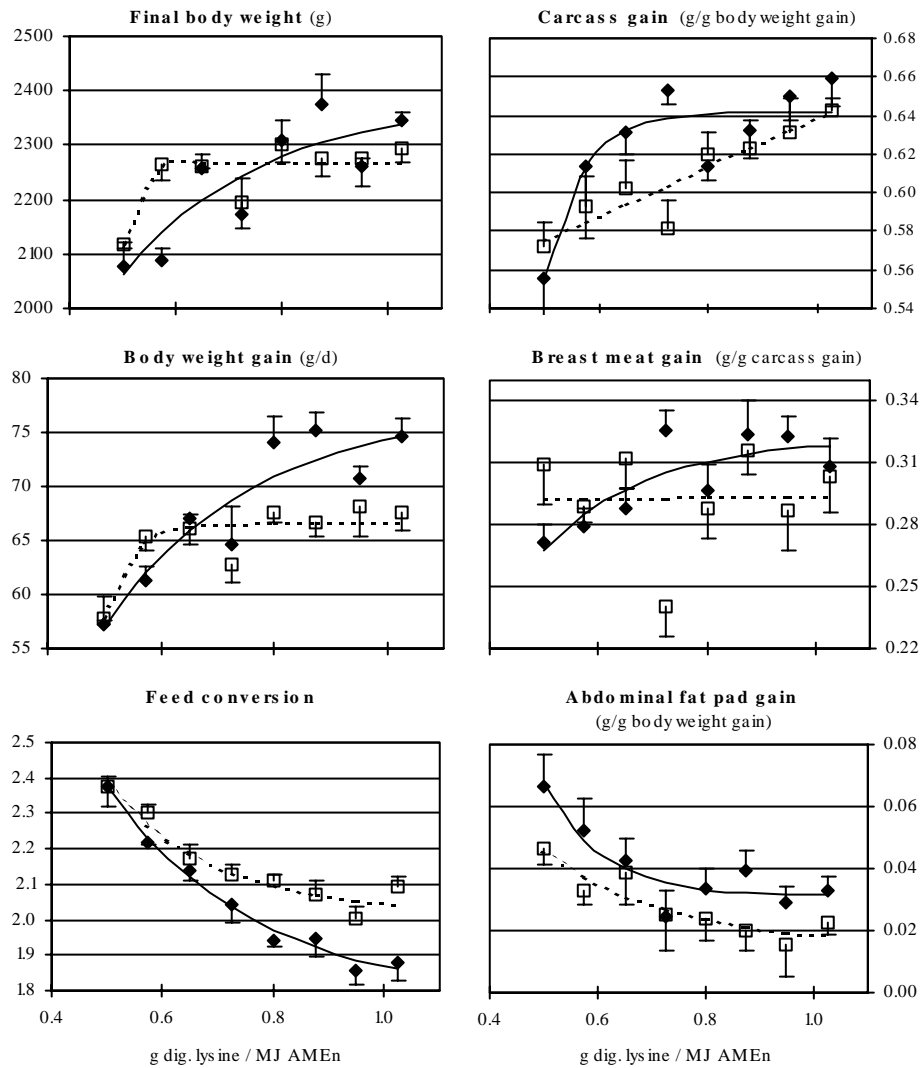


Figure 2. Dose-related responses of male broiler chickens to dietary balanced protein to energy ratio (BPE-ratio) in EXP2 (26-41 d of age). High (---, □) and low (—, ◆) BPE-ratio in EXP1 (11-26 d of age). Values are averages with *E* indicated by the error bars (*n*=3).

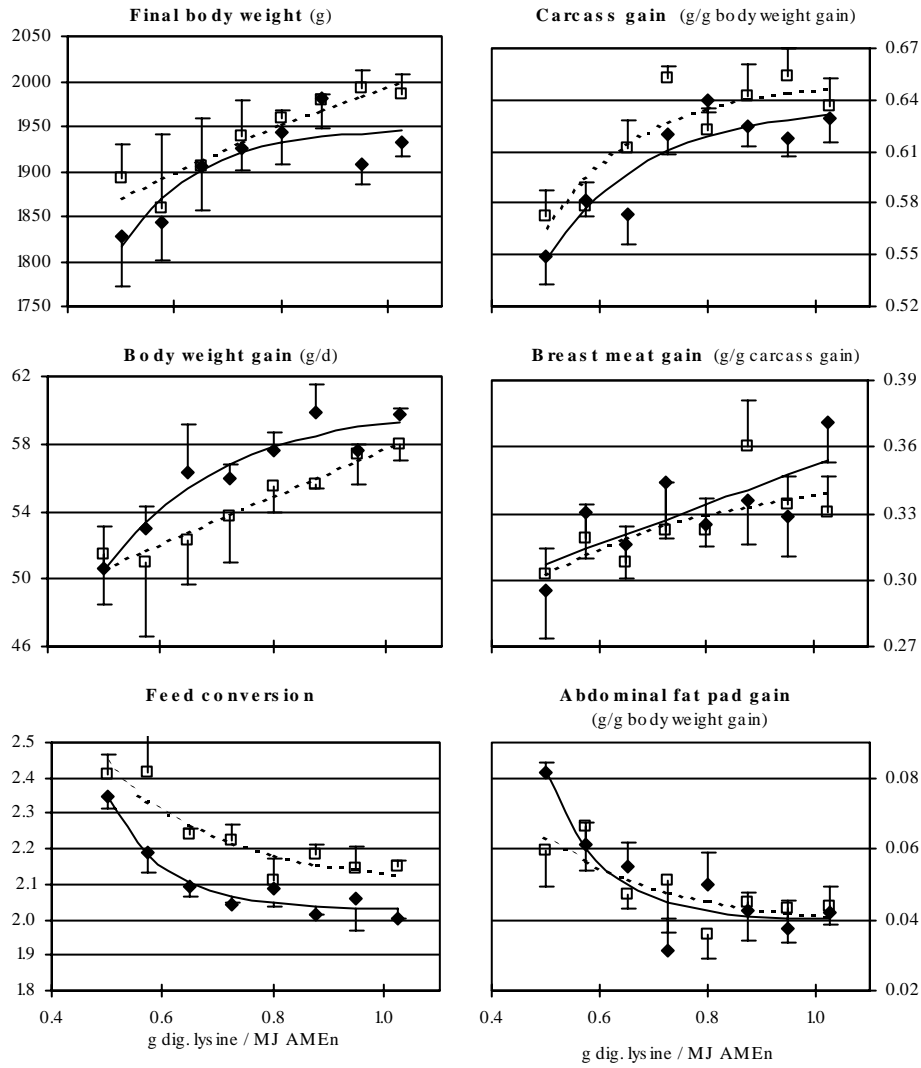


Figure 3. Dose-related responses of female broiler chickens to dietary balanced protein to energy ratio (BPE-ratio) in EXP2 (26-41 d of age). High (---, □) and low (—, ◆) BPE-ratio in EXP1 (11-26 d of age). Values are averages with SE indicated by the error bars (n=3).

It was therefore possible to use linear regression to estimate the overall effect of diet in EXP2 on pericardial moisture score. This latter analysis revealed that, although the amount of variance explained was rather low ($R^2 = 0.10$), pericardial moisture score increased significantly ($P < 0.01$) by 0.695 (SE 0.22) for each additional gram digestible lysine per MJ AMEn in EXP2. Over the range of BPE ratios used the pericardial moisture score would be predicted to increase from 0.25 to 0.61.

Thus, diet composition at early ages (11 to 26 d of age) did not affect the ascites-related metabolic variables at 41 d, but it did affect mortality during the 26 to 41 d period.

DISCUSSION

Statistical model

The response of growing broiler chickens to protein level in the diet is characterised by diminishing increments of response as the dietary protein level increases, up to levels where a plateau in output is reached (for example, Clark *et al.*, 1982; Smith *et al.*, 1998). The exponential model used in this study has these properties and could be an appropriate one to describe such responses. The model has the additional advantage that its parameters have biological meanings. Parameters a , k , and c represent respectively, the performance level at a fixed low BPE ratio (0.575 g/MJ), the maximum performance level (asymptote) and the rate of increase in performance from a to k when BPE ratio is increased above 0.575 g/MJ.

Careful interpretation is needed when the estimated asymptote for the exponential model is much higher than the highest observed response in the test. An extreme example concerns the females in EXP2, fed on the high protein diet in EXP1. The estimated asymptote in body weight is 2197 g (Table 4), whereas the highest observed body weight in this treatment group is about 1990 g (Figure 3). Consequently, the precision of the estimated asymptote is very low (SE is 744 g; Table 4). Within the range of protein levels tested, however, the exponential response model gives in all cases an accurate description of the data in this experiment. This conclusion is based on the random distribution of the data along the fitted response curves (Figures 1 to 3). The flexibility of the model, particularly the flexibility in curve steepness, contributed to the goodness of fit.

Feed intake

Responses in performance of broilers that have unrestricted access to diets with varying concentrations of a given nutrient reflect a combination of two possible effects. Changing the dietary content of a nutrient may change feed intake *per se* (thus affecting the absolute intake of the nutrient). Secondly, it can also change the metabolic efficiency by which the nutrient is utilised (Baker, 1984). An attempt to distinguish between these two effects is outside the scope of this paper. However, data on the average feed intakes at different treatments groups are given (Table 5) to allow such an analysis to be made.

Age period

The present study was designed to study the carry-over effects of feeding a low protein diet from 11 to 26 d of age on broiler responses to protein level in the diet during 26 to 41 d of age. Part of the interest was in nutritional carry-over effects on metabolic disorders. Scheele *et al.* (1999) found that feeding broilers a diet low in first limiting amino acids to energy ratio in the early growing phase (14 to 21 d of age) may lower the incidence of metabolic disorders at later age. The same age period, including a safety margin of some additional d was therefore chosen as the restriction period for the present study. The period of 26 to 41 d was chosen as the re-alimentation period where d 41 was supposed to be the average slaughter age in practice. Body composition changes in this period are relevant for carcass quality.

Nutritional carry-over effects on growth and development

Results of our study indicate that broiler responses to dietary balanced protein level in a later grower phase depend on the balanced protein level in the diet that was fed previously. Such carry-over effects were found with growth rate and feed conversion, and to a less extent with composition of the gain. Compensatory growth and improved feed utilisation after protein restriction, as found in the present study, have been demonstrated before with turkeys (Auckland and Morris, 1971) and with broilers (Moran, 1979; Pesti and Fletcher, 1984).

In studies on compensatory growth, it is common practice to compare the performance of previous restricted animals with that of control animals that are supposed to show unrestricted growth throughout (for example, Plavnik and Hurwitz, 1989; Zubair, 1994). In the present study, growth

rates during the re-alimentation period were up to 25% lower than breeder standards, for the restricted birds as well as for the control group. This raises the question of whether the overall reduction in growth rate may have affected the possibility for the restricted birds to show compensatory growth, that is, to grow faster than the control birds. Marks (1978) found that fast growing quail lines were capable of demonstrating greater compensatory growth following a protein restriction than non-selected control quail. Cherry *et al.* (1978) noted the opposite effect of genetics on compensatory growth capacity. The slower growing broiler strains in their study exhibited more compensatory gain than the faster grower strains. Neither study proves, however, that the suggested correlation between overall growth rate and capacity for growth compensation between strains also applies for a given genotype.

No evidence was found in the present study for a nutritional carry-over effect of previous feeding on the proportion of carcass in the gain. Similarly Moran (1979) and Pesti and Fletcher (1984) found no effect of dietary protein level on carcass yield, neither immediately after feeding a low protein diet, nor after a re-alimentation period of 1 or 2 weeks, respectively. This suggests that in their studies, as in the study reported here, the proportion of carcass in the gain during compensation was not affected by the early protein limitation.

Composition of the gain in the present study was not completely independent of previous nutrition. The proportion of abdominal fat in the gain in males, for example, was increased following the early protein restriction. This higher gain of abdominal fat was attended by a lower feed conversion ratio. Due to its relatively low water content and high fat content, adipose tissue requires more energy per kg of gain than lean tissues. The mentioned carry-over effects for abdominal fat and feed conversion in males seem therefore contradictory. It is possible that the response in accretion of total body fat was opposite to the response in abdominal fat. Data published by Pesti and Fletcher (1984) indicate indeed that, during the week that followed the period with inadequate protein nutrition, feed conversion improved, proportion of abdominal fat in the gain increased whereas total fat content in the gain decreased. The abdominal fat pad is often used as an indicator for carcass fat, including studies on compensatory growth (for example, Plavnik and Hurwitz, 1988, 1989; Zubair, 1994). However, for the latter type of studies, gain in abdominal fat pad may be not a reliable indicator of gain in carcass fat or total body fat.

Table 5. Effects of dietary BPE-ratio¹ on average daily feed intake (g/d) with broiler chickens in EXP1 and EXP2

Experiment	Sex	BPE-ratio in EXP1 ³	BPE-ratio											P ⁴
			0.500	0.575	0.650	0.725	0.800	0.875	0.950	1.025	1.100			
EXP1	Male		97.7	95.9	95.7	94.3	91.6	91.5	92.3	93.7	**			
	Female		84.2	80.7	82.6	80.5	81.5	83.2	82.4	81.4	NS			
EXP2	Male	Low	135.6	135.8	143.4	131.9	143.7	146.0	131.4	140.3	**			
		High	137.2	150.4	143.4	133.4	142.7	137.8	136.4	141.1	**			
	Female	Low	118.6	116.3	117.7	114.4	120.3	120.7	118.6	119.8	NS			
		High	123.6	122.2	117.2	119.2	116.9	121.4	123.0	124.5	NS			

1 Balanced protein to energy ratio (g digestible lysine per MJ AMEn).

2 EXP1: 11-26 d of age; EXP2: 26-41 d of age.

3 Low and high represent 0.725 and 1.025 g digestible lysine per MJ AMEn, respectively, in the diet during EXP1.

4 Level of significance for test if feed intake is affected by BPE-ratio; NS: $P > 0.10$; * $0.10 > P > 0.05$; ** $0.05 > P > 0.01$; *** $P < 0.01$.

The water content of the carcass, relative to the protein content, may be reduced in broilers chickens by inadequate protein nutrition (Chapter 2). Possible compensation of the water content of the carcass after the period of protein restriction is another factor that could explain the efficient feed conversion during re-alimentation.

The abdominal fat pad is an important organ for the storage of energy in poultry. The high relative gain in abdominal fat with the males following the period of inadequate protein nutrition may reflect a metabolic adaptation induced by a period of inadequate nutrition in general. Following a quantitative feed restriction and refeeding, for example, Beane *et al.* (1979) found, besides an improvement in feed efficiency, a significant increase in the relative weight of the abdominal fat of male broilers.

Nutritional responses in growth of the abdominal fat pad may be different between sexes. In the mentioned study of Beane *et al.* (1979), no effect of feed restriction was found on abdominal fat weight after refeeding in females. Likewise, in the present study proportion of abdominal fat pad in the gain following feed restriction was increased in males, but not affected in females. The hypothesis that the nutritional carry-over effects would depend on the sex of the birds was not only confirmed for abdominal fat, but also for body weight gain. Plavnik and Hurwitz (1988) found male broilers to have a greater ability to exhibit compensatory growth after a period of undernutrition than females. The same conclusion can be drawn from our study, but only if the crude protein level in the re-alimentation diet is above 20% (Figures 2 and 3).

Protein requirements during re-alimentation

A requirement for protein can be seen as the minimum level of protein in the diet at which some measure of output (for example, weight gain) is optimised. Feeding a low protein diet to growing turkeys or broilers during a certain age period may increase the protein requirement for optimal performance in the subsequent age period (Auckland and Morris, 1971; Plavnik and Hurwitz, 1989). Only males were used in these two studies. Also with the males in the present study, protein requirement to optimise weight gain following a period of protein restriction was clearly increased compared to unrestricted controls (Figure 2). For the females, and for the other measures of output (for example, feed conversion ratio or breast

meat gain), differences in protein requirement due to previous protein restriction were much less pronounced than for weight gain of the males (Figure 2). The higher ability for compensatory growth of males compared to females, as suggested earlier, appears to have consequences for the protein requirement during growth compensation.

Nutritional carry-over effects on health status

Scheele *et al.* (1999) found that the effect of feeding strategy on metabolic disorders occurred only in sensitive birds – the males of a pure line with low ascites resistance. The genotype used in our experiment was less sensitive to metabolic disorders than the sensitive strain used by Scheele *et al.* (1999). The birds used by Scheele *et al.* (1999) were also housed under the ascites-provoking conditions of low environmental temperature. In our study, temperature was intended to be optimal for broiler health. The differences in genotype and environment between Scheele *et al.* (1999) and our study may explain why the incidence of metabolic disorders in our study was not decreased by feeding a low protein diet at an early age.

Feeding a low instead of a high protein diet in EXP1 increased mortality in EXP2. This may be related to the compensatory gain of these broilers in EXP2. Fast growth, particularly in combination with a low feed conversion, is known to be a risk factor for metabolic disorders (Scheele *et al.*, 1991; Julian, 1993). However, rapid gain like the compensatory growth observed here, may also be a risk for non-metabolic illnesses like skeletal disorders (Leeson *et al.*, 1995). Such negative effects of compensatory gain on the health status may overrule the beneficial effects of a low protein diet at early age, particularly if the genetic strain is insensitive to metabolic disorders. The sensitivity of a broiler strain for metabolic disorders needs to be considered before a low protein diet for the early growing phase is included in feeding programs intended to promote health.

Conclusions

The present study has demonstrated that responses in broiler performance and body composition to dietary balanced protein to energy ratio (BPE ratio) depend on previous protein nutrition and sex. Up to 10% improvement in both weight gain and feed conversion at later age (EXP2) were observed if BPE ratio during the early growing phase (EXP1) was decreased. Effects of BPE ratio during EXP1 on the incidence of metabolic disorders in EXP2 were not observed. The lower BPE ratio in EXP1 increased mortality in EXP2 from 0.8 to 3.6%. The results strongly suggest

that protein levels in grower and finisher diets should not be optimised independently, but simultaneously.

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Chapter 4

**Dietary Balanced Protein in Broiler Chickens: a Flexible
and Practical Tool to Predict Dose-Response Curves**

Paper by: R. M. Eits, R. P. Kwakkel, M. W. A. Verstegen and L A. den Hartog. Submitted to British Poultry Science.

ABSTRACT An exponential dose-response model was developed that predicts growth rate, feed conversion and carcass and breast meat yield of broilers as a function of dietary balanced protein level (DBP). The model was developed to serve as a tool for nutritionists. The model helps determining the DBP that maximise profit. The model avoids practical disadvantages of existing methods. In contrast with mechanistic models, only data that are generally known by broiler nutritionists are required as input. Compared to predictions derived from one or a few feeding trials, the model predictions are probably more flexible and accurate for a wider range of genotypes because a description of the type of broiler was included as input for the model.

Broiler response studies from literature and the Nutreco Poultry Research Centre (27 data sets in total) were used in the model development to select significant variables, to quantify the parameters and to evaluate the accuracy of the predictions. Input variables were DBP, asymptotic performance level, age, year (indicating genetic potential) and sex. The model, including the assumption that the shape of the dose-response relationships to DBP is a constant gave an accurate simulation of growth rate, feed conversion and breast meat for nearly all data sets. Accuracy was less for carcass. In Chapter 5, the developed model is used to evaluate how profitability of a broiler enterprise depends on DBP.

INTRODUCTION

Dietary protein level has a considerable impact on growth rate, feed efficiency and body composition of broiler chickens (Skinner *et al.*, 1991; Smith *et al.*, 1998), but also on the cost price of the diet. Thereby, protein level of the diet strongly affects costs as well as revenues in broiler meat production. The decision of what protein level is optimal in broiler diets depends on the desired quality of the end product and is also an economic question. Calculation of the economic optimal protein level is problematic, particularly because an accurate quantitative description is needed on how broiler performance, for example growth rate, feed conversion and body composition, relates to dietary protein level.

Such dose-response relationships are often empirically quantified in a feeding experiment (Smith *et al.*, 1998) or sometimes predicted by means of a mechanistic simulation model (Gous, 1998). Both approaches are of limited value for nutritionists to determine economic optimal protein levels. Empirically determined relationships require expensive, time-consuming testing and are only reliable under the exact conditions (in terms of bird characteristics, feed and environment) of the specific experiment. Mechanistic simulation models have the disadvantage that often complex input data are required that are unknown by most broiler nutritionists.

The objective of this study was to develop an alternative method to generate dose-response curves for modern broiler genotypes under commercial conditions. It predicts growth rate, feed conversion and carcass and breast meat yield at various dietary protein levels. The new method avoids the above-mentioned disadvantages of the existing methods. For example, some input data are used to describe the type of broiler in order to make the predictions flexible and accurate for a wider range of genotypes than predictions derived from one or a few empirical feeding trials. In addition, and in contrast to a mechanistic model, only data that are generally known by broiler nutritionists are required as input.

MATERIALS AND METHODS

The objective of this study was realised in two steps. First, a mathematical model was developed that meets the imposed criteria as formulated in the

objective. Secondly, experimental data were applied to further develop this model (selection of variables and quantification of parameters) and to evaluate the accuracy.

Development of the mathematical model

Dose-response relationships obtained in different broiler studies may be strongly dissimilar due to differences related to the bird, the feed and the environment. Dose-response relationships to dietary protein may differ in three aspects, as is shown in Figure 1. The three aspects are (1) the broiler performance at a quite high protein level (indicating the asymptotic performance level), (2) the protein level needed to obtain a given response (horizontal position of 'intercept') and (3) the rate of response to increasing protein levels (shape of the response curve). This implies that, if a nutritionist has information about these three sources of variation for the broilers in his integration, dose-response relationships can be constructed without actual experimentation.

The mathematical model for this study should represent the three sources of variation in response curves identified above (asymptote, intercept and shape of the curve). Secondly, it should allow for the typical shape of response of growing broiler chickens to dietary protein, which is characterised by diminishing increments of response as the dietary protein level increases, up to levels where a plateau in output is reached (Clark *et al.*, 1982). The following exponential equation meets these criteria and was therefore chosen as the mathematical model for the prediction of dose-response curves:

$$GR = B + (A - B) * (1 - e^{-C*(Lys-D)}) \quad [1]$$

where: GR = growth rate (g/d); B = broiler performance ('intercept') at a fixed Lys level (D); A = asymptotic performance level; C = shape of response to Lys level; Lys = dietary Lys level (g Lys/kg diet), used as a reference for balanced protein level and D = fixed level of Lys. Model [1] has been used previously to describe the responses of turkeys to dietary Lys (Lehmann *et al.*, 1996) and of broiler chickens to dietary balanced protein (Chapter 3).

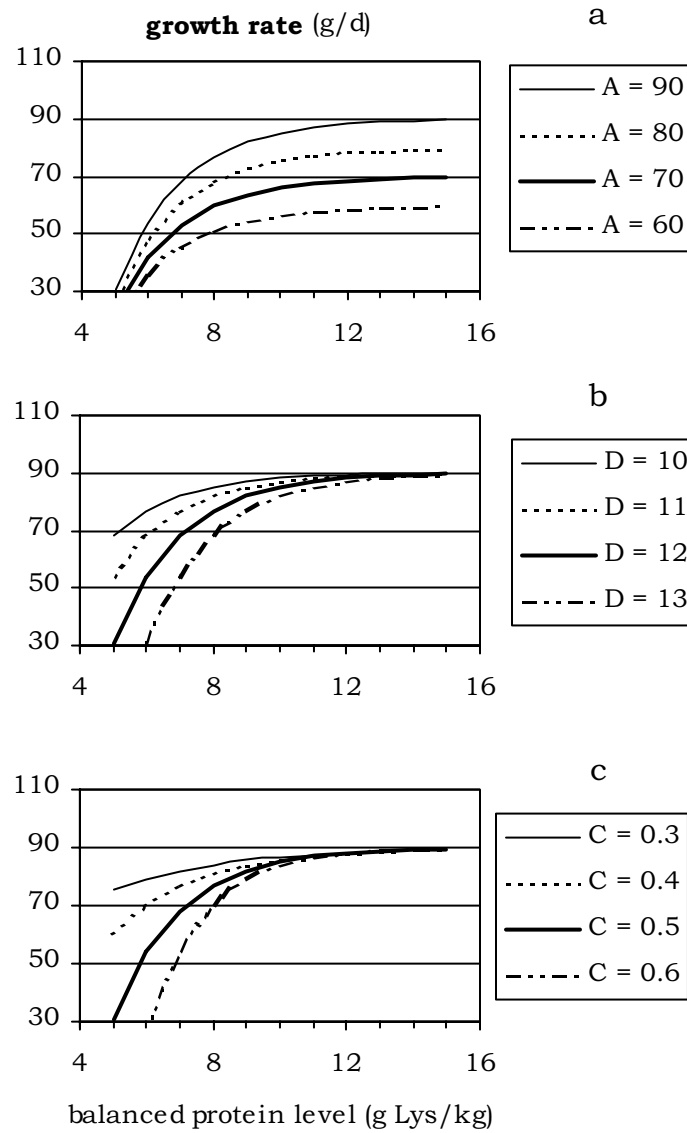


Figure 1. Three sources of variation in dose-response curves: vertical position (panel a), horizontal position (panel b) and shape (panel c). Hypothetical dose-response curves based on model [2]: $GR = 0.98 \cdot A + 0.02 \cdot A \cdot (1 - \exp(-C \cdot (Lys - D)))$; see text.

The asymptotic performance level is usually reasonably well-known by an experienced nutritionist. Information about the intercept and the shape of the response curve may be less straightforward and is discussed below.

The intercept was defined as the Lys level (D) that is required for a given growth rate B (model [1]). Nutritionists have more knowledge about growth rates at practical, high Lys levels than at low Lys levels. Therefore, to maximise the reliability of the estimated intercept (D), the intercept was defined at a relatively high Lys level; namely the Lys level at which growth rate (B) is at 98% of the asymptote (A). This somewhat arbitrarily chosen Lys level will be referred to as Lys requirement. Model [1] thus becomes:

$$GR = 0.98 * A + 0.02 * A * \left(1 - e^{-C*(Lys-D)}\right) \quad [2]$$

where, additionally to [1]: D represents Lys requirement (g Lys / kg diet). It should be noted that this model predicts growth rates below $0.98*A$ (if Lys $< D$) as well as above $0.98*A$ (if Lys $> D$).

To further improve the reliability of the estimated Lys requirement (D), a model was developed to predict D from other variables. The variables should represent major sources of variation in D . Moreover, in order to be of practical relevance, only those variables should be used of which broiler nutritionists know the actual value under field conditions. Variables that are in accordance with those criteria are age, sex and year. Year represents the genetic potential of the broiler (for example, 2002). Experimental data proves that D may be affected by age and sex (Smith *et al.*, 1998; Chapter 3) as well as year (genetic potential) (Whitehead, 1990; Leclercq and Guy, 1991; Smith *et al.*, 1998). Assuming that D is a linear function of age, sex and year, model [2] becomes:

$$GR = 0.98 * A + 0.02 * A * \left(1 - e^{-C*(Lys-[E+F*age+G*year+H*sex])}\right) \quad [3]$$

where: age is the average day of the relevant age period (d) and E , F , G and H are model parameters that relate age, year and sex to Lys requirement (D). Values for sex are 1 (males), 0 (females) and 0.5 (as hatched). If a nutritionist has data on Lys requirement under his field conditions that he believes to be more accurate than the general predictions based on age, sex and year, then he can use his own estimate for Lys requirement to predict the dose-response curves.

The shape of the response curve (C) is difficult to quantify under field conditions. It was assumed that C is similar for laboratory conditions and

field conditions. In addition, it was assumed that C is independent of broiler characteristics (for example, age, sex, breed) and feed characteristics (for example, dietary energy level).

For feed conversion ratio (FCR), dose response curves can be predicted using the same method as for growth rate. An adjustment was needed, because FCR minimises with increasing protein level whereas growth rate maximises. Lys requirement for FCR was therefore defined as the Lys level at which FCR is at 102% of the asymptote. Thus, in analogy to model [3], the model for FCR becomes:

$$FCR = 1.02 * A - 0.02 * A * \left(1 - e^{-C*(Lys-[E+F*age+G*year+H*sex])}\right) \quad [4]$$

Similar models, but without the variable year (see selection of variables), were used to simulate dose response curves for carcass yield (g/g broiler weight) and breast meat yield (g/g carcass):

$$Carcass = 0.98 * A + 0.02 * A * \left(1 - e^{-C*(Lys-[E+F*age+H*sex])}\right) \quad [5]$$

$$Breast\ meat = 0.98 * A + 0.02 * A * \left(1 - e^{-C*(Lys-[E+F*age+H*sex])}\right) \quad [6]$$

Selection of data sets

Studies on broiler responses to dietary balanced protein level, either from literature or from the Nutreco Poultry Research Centre, were used to further develop the model and to evaluate its accuracy. The following selection criteria were applied to select appropriate data sets. Growing broiler chickens of a commercial line must have been fed *ad libitum* with isoenergetic diets containing varying amounts of Lys. Preferably, there should have been at least five levels of Lys in each test in order to get a reliable estimate of the shape of the dose-response curve (Remmenga *et al.*, 1997). One study with only four Lys levels (Surisdiarto and Farrell, 1991) was accepted, however, as that study included scarce data on low Lys levels in combination with young broilers. The minimum amounts of essential amino acids relative to Lys should have been constant among treatments to keep the right balance between amino acids (balanced protein); not necessarily the same balance among all studies. As the response of rapidly developing broiler chickens changes from day to day, the experimental period ought to have been no longer than 21 d. Otherwise, the response would reflect an accumulation of too much physiological different growth phases. An overview of the studies that satisfied the selection criteria is given in Table 1.

TABLE 1. Data sets used for the model construction ¹

Reference ²	Data set	Age period (d)	Breed ³	Details
1. Clark <i>et al.</i> , 1982	1	7 – 21	Hybro	As hatched; EXP1; dietary energy on NE-basis
2. Surisdiarto and Farrell, 1991 ⁴	1	0 – 21	See 'details'	Males, commercial strain (Australian Poultry Ltd)
3. Skinner <i>et al.</i> , 1991	1	42 – 49	Cobb 500	As hatched, average results over 6 similar trials
4. Skinner <i>et al.</i> , 1992	1, 2	42 – 49	Cobb 500	Data sets 1 (males) and 2 (females); Lys 1.08 g/kg during 0-42 d
- idem	3, 4	42 – 49	Cobb 500	Data sets 3 (males) and 4 (females); Lys 1.22 g/kg during 0-42 d
5. Smith <i>et al.</i> , 1998 ⁵	1, 2	18 – 32	See 'details'	Data sets 1 (Ross*Ross 208) and 2 (Peterson*Arbor Acres)
6. Eits, 2001 (unpublished)	1, 2	14 – 35	Hybro-MPN	Data sets 1 (males) and 2 (females)
- idem	3, 4	14 – 35	Hybro-SK	Data sets 3 (males) and 4 (females)
- idem	5, 6	35 – 49	Hybro-MPN	Data sets 5 (males) and 6 (females)
- idem	7, 8	35 – 49	Hybro-SK	Data sets 7 (males) and 8 (females)
7. Eits <i>et al.</i> , 2003 ⁶	1, 2	11 – 26	Hybro-G	Data sets 1 (males) and 2 (females)
- idem	3, 4	26 – 41	Hybro-G	Data sets 3 (males) and 4 (females); Lys 1.04 g/kg during 11-26 d
- idem	5, 6	26 – 41	Hybro-G	Data sets 5 (males) and 6 (females); Lys 1.42 g/kg during 11-26 d
8. Weurding, 2002 ⁷	1, 2	9 – 18	Cobb 500	As hatched, data sets 1 (peas in diet) and 2 (tapioca in diet)
9. Eits, 2002 (unpublished)	1, 2	21 – 42	Hybro-G	Data sets 1 (males) and 2 (females)

¹ Carcass and breast meat data were only available from references 4, 5, 6, 7 and 9.

² Smith *et al.* (1998) used crumbles and Clark *et al.* (1982) used mash feed, whereas the experimental diets in all other studies were pelleted.

³ Hybro-MPN and Hybro-SK have similar growth rates and feed conversion, but Hybro-SK has a much higher breast meat yield than Hybro-MPN.

⁴ 5 Lys levels at each of 4 ideal protein levels were tested (Exp2). Data of diets with the lowest Lys level at each ideal protein level were used in the present study.

⁵ Smith *et al.* (1998) reported no Lys and digestible Lys levels for the diets. However, reliable estimates for total and digestible Lys could be derived from the raw material composition and CP levels of the diets, using tabulated values (CVB, 2000), because corn, soy bean meal and synthetic amino acids were the only amino acid sources in their diets.

⁶ AME level was not completely constant among treatments but decreased gradually with about 6% from the lowest to the highest protein level.

⁷ Weurding (2002) only published digestible Lys levels, but total Lys levels were supplied upon request.

Selection of variables and quantification of parameters

The collected data sets were used for the selection of significant variables to predict Lys requirement (age, year, sex) in the model for growth rate. At the same time, the model parameters (C , E , F , G , H ; see model [3]) were quantified. This was done by fitting model [3] to all the selected data sets (Table 1) simultaneously, using non-linear regression (SPSS, 1999).

The selection of the variables to predict Lys requirement was done by running the data analysis several times with the variables age, sex and year being added one by one to the basis model. The basis model is the model with dietary Lys level and A as the only variables (see also Table 2). Regarding the order of adding the variables, priority was always given to the variable that gave the largest single improvement in explained variance. The significance of improvement of the model by inclusion of an additional variable was evaluated by means of an F-test.

For this data analysis, variable A (asymptotic growth rate) in model [3] had a predetermined value, which was different for each data set. This A -value was calculated as the average growth rate at the highest two Lys levels in that data set. Parameter C in model [3] was quantified by means of non-linear regression, similarly as for parameters E , F , G and H . Thus, parameter C in the model for growth rate had the same value for all data sets. The year of publication of an experiment was taken as the value for the variable year. Details on the statistics are given below. Lys requirements for FCR, carcass yield and breast meat yield were modelled in the same way as for growth rate, using models [4]-[6] instead of model [3].

The model was constructed with total or digestible Lys as input variable. For 20 data sets (references 5, 6, 7, 8 and 9 in Table 1) out of 27 in total, Lys digestibility figures were available. Only these 20 data sets were used for the development of the model with digestible Lys. The range of years among these data sets is relatively small (1998-2002). Therefore, year (=genetic potential) was not included in the model with digestible Lys. For the same reason, year was not included in the models for carcass and breast meat yield.

Regression models were fitted to the treatment averages of the experimental data. Non-linear (Levenberg-Marquardt algorithm; Moré, 1977) regression procedures (SPSS, 1999) were used to analyse the data. The significance of improvement of a model by inclusion of an additional variable was assessed

by an F-test: the difference in the residual sum of squares between the model with and the model without the additional variable, divided by the difference in degrees of freedom of the residuals, was tested against the residual mean square of the model with the additional variable.

Accuracy of the model

The accuracy of the model predictions was tested by plotting the experimental data and the predicted curves together in a graph, with a separate graph for each data set. Model predictions came from models [3]-[6]. A good fit, that is a random distribution of the experimental data around the predicted dose response curves, would signify a good accuracy of the model. The goodness-of-fit should be evaluated at the lower Lys levels in each data set. The simulation close to the asymptote is accurate by definition, as the asymptote was defined as the average growth rate or FCR at the highest two Lys levels in a data set.

A separate graph for each data set makes it possible to judge the goodness-of-fit for each data set. In this way, possible deviations in the model predictions for one or more data sets could be traced back to factors that may explain them (for example, breed, sex or others; Table 1).

RESULTS

Selection of variables and quantification of parameters

Growth rate The models developed for growth rate and the significance of the variables (age, year, sex) to predict Lys requirement for maximising growth rate are presented in Table 2. The basis model, with total Lys and *A* as the only variables, explained already 96.1% of the variation (model 3.1 in Table 2). Age and year were the first and second most important variables to predict Lys requirement, and together ($R^2 = 97.9\%$) they explained about half of the variance that was not explained by the basis model. Sex did not give a significant improvement in the model ($P > 0.10$; Table 2).

For the model with digestible Lys, the significance of the variables was similar as for the model with total Lys (Table 2). The R^2 for the best model with digestible Lys (95.4%) was lower than for the best model with total Lys (97.9%).

Table 2. An exponential dose response model¹ predicting growth rate (g/d) of broiler chickens: Significance of the variables to model Lys requirement and estimated values of the parameters (asymptotic SE between brackets)

Lys	Variables to model Lys requirement ²	Latest added variable	R ² _{adj}	P ³	C	E	F	G	H
Total	3.1 -	-	96.1		.316 (.04)	11.2 (.59)	-	-	-
	3.2 age	Age	97.7	***	.438 (.03)	15.1 (.53)	-.127 (.012)	-	-
	3.3 age, year	Year	97.9	***	.610 (.05)	-173.2 (30)	-.0999 (.009)	.0935 (.015)	-
	3.4 age, year, sex	Sex	97.9	NS	.610 (.05)	-172.9 (30)	-.0999 (.009)	.0933 (.015)	-.024 (.18)
Digestible ⁴	3.1 -	-	94.8		.757 (.08)	9.14 (.23)	-	-	-
	3.2 age	Age	95.4	***	.759 (.07)	10.8 (.45)	-.0564 (.013)	-	-
	3.5 age, sex	Sex	95.4	NS	.756 (.07)	10.7 (.47)	-.0561 (.014)	-	.063 (.20)

1 $GR = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + C^*year + H^*sex])))$, where: GR = growth rate (g/d); A = asymptotic growth rate (g/d); C = shape of the dose-response curve; Lys = dietary Lys level (g/kg); [...] = Lys requirement' (dietary Lys level at which $GR = 0.98^*A$), as a function of broiler age (d), year (e.g. 2002), and sex (1 = males; 0 = females; 0.5 = as hatched).

2 Variables to model Lys requirement were added to the model for growth rate (see footnote 1) one-by-one.

Model 3.1: $GR = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E])))$, basis model; for meaning of the variables and parameters, see footnote 1.

Model 3.2: $GR = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age])))$; for meaning of the variables and parameters, see footnote 1.

Model 3.3: $GR = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + G^*year])))$; for meaning of the variables and parameters, see footnote 1.

Model 3.4: $GR = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + G^*year + H^*sex])))$; for meaning of the variables and parameters, see footnote 1.

Model 3.5: $GR = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + H^*sex])))$; for meaning of the variables and parameters, see footnote 1.

3 Significance of the contribution to the model by the latest added variable; NS: $P > 0.10$; * $0.10 > P > 0.05$; ** $0.05 > P > 0.01$; *** $P < 0.01$.

4 For the model on digestible Lys, a selection of 20 out of the 27 data sets in total was used (see text).

Table 3. An exponential dose response model¹ predicting feed conversion ratio (FCR) of broiler chickens: Significance of the variables to model Lys requirement and estimated values of the parameters (asymptotic SE between brackets)

Lys	Variables to model Lys requirement ²	Latest added variable	R ² _{adj}	P ³	C	E	F	G	H
Total	4.1 -		83.3		.413 (.05)	11.9 (.64)	-	-	-
	4.2 age	Age	96.4	***	.462 (.02)	15.2 (.36)	-.104 (.006)	-	-
	4.3 age, year	Year	97.0	***	.540 (.02)	-99.7 (16)	-.0941 (.005)	.0572 (.008)	-
	4.4 age, year, sex	Sex	97.0	NS	.538 (.02)	-97.8 (17)	-.0936 (.005)	.0562 (.008)	.0679 (.13)
Digestible ⁴	4.1 -		96.1		.539 (.03)	10.7 (.22)	-	-	-
	4.2 age	Age	96.9	***	.567 (.03)	12.2 (.33)	-.0562 (.009)	-	-
	4.5 age, sex	Sex	97.5	***	.544 (.03)	11.7 (.32)	-.0467 (.008)	-	.716 (.13)

1 FCR = 1.02*A - 0.02*A*(1 - exp(-C*(Lys - [E + F*age + G*year + H*sex]))) , where: FCR = feed conversion ratio; A = asymptotic FCR; C = shape of the dose-response curve; Lys = dietary Lys level (g/kg); [..] = Lys 'requirement' (dietary Lys level at which FCR = 1.02*A), as a function of broiler age (d), year (e.g. 2002), and sex (1 = males; 0 = females; 0.5 = as hatched).

2 Variables to model Lys requirement were added to the model for FCR (see footnote 1) one-by-one.

Model 4.1: FCR = 1.02*A - 0.02*A*(1 - exp(-C*(Lys - [E]))) , basis model; for meaning of the variables and parameters, see footnote 1.

Model 4.2: FCR = 1.02*A - 0.02*A*(1 - exp(-C*(Lys - [E + F*age]))) , for meaning of the variables and parameters, see footnote 1.

Model 4.3: FCR = 1.02*A - 0.02*A*(1 - exp(-C*(Lys - [E + F*age + G*year]))) , for meaning of the variables and parameters, see footnote 1.

Model 4.4: FCR = 1.02*A - 0.02*A*(1 - exp(-C*(Lys - [E + F*age + G*year + H*sex]))) , for meaning of the variables and parameters, see footnote 1.

Model 4.5: FCR = 1.02*A - 0.02*A*(1 - exp(-C*(Lys - [E + F*age + H*sex]))) , for meaning of the variables and parameters, see footnote 1.

3 Significance of the contribution to the model by the latest added variable; NS: P > 0.10; * 0.10 > P > 0.05; ** 0.05 > P > 0.01; *** P < 0.01.

4 For the model on digestible Lys, a selection of 20 out of the 27 data sets in total was used (see text).

Table 4. An exponential dose response model¹ predicting carcass yield (g/g body weight) of broiler chickens: Significance of the variables to model Lys requirement and estimated values of the parameters (asymptotic SE between brackets)

Lys	Variables to model Lys requirement ²	Latest added variable	R ² _{adj}	P ³	C	E	F	G	H
Total	4.1 -	-	97.7		.137 (.11)	-.05 (7.9)	-	-	-
	4.2 age	Age	98.0	***	.406 (.12)	16.2 (2.3)	-.310 (.099)	-	-
	4.3 age, sex	Sex	98.1	**	.531 (.13)	15.6 (1.5)	-.251 (.06)	-	-1.52 (.70)
Digestible ⁴	4.1 -	-	98.2		.786 (.16)	7.1 (.2)	-	-	-
	4.4 sex	Sex	98.3	**	.945 (.18)	7.5 (.2)	-	-	-.65 (.31)
	4.3 sex, age	Age	98.3	**	.947 (.19)	9.5 (.6)	-.061 (.02)	-	-1.24 (.38)

1 Carcass = $0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + C^*year + H^*sex])))$, where: Carcass = carcass yield (g/g body weight); A = asymptotic carcass yield (g/g); C = shape of the dose-response curve; Lys = dietary Lys level (g/kg); [...] = Lys 'requirement' (dietary Lys level at which Carcass = 0.98^*A), as a function of broiler age (d), year (e.g. 2002), and sex (1 = males; 0 = females; 0.5 = as hatched).

2 Variables to model Lys requirement were added to the model for carcass yield (see footnote 1) one-by-one.

Model 4.1: Carcass = $0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E])))$, basis model; for meaning of the variables and parameters, see footnote 1.

Model 4.2: Carcass = $0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age])))$; for meaning of the variables and parameters, see footnote 1.

Model 4.3: Carcass = $0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + H^*sex])))$; for meaning of the variables and parameters, see footnote 1.

Model 4.4: Carcass = $0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + H^*sex])))$; for meaning of the variables and parameters, see footnote 1.

3 Significance of the contribution to the model by the latest added variable; NS: $P > 0.10$; * $0.10 > P > 0.05$; ** $0.05 > P > 0.01$; *** $P < 0.01$.

4 For the model on digestible Lys, a selection of 18 out of the 22 data sets in total was used (see text).

Table 5. An exponential dose response model¹ predicting breast meat yield (g/g carcass) of broiler chickens: Significance of the variables to model Lys requirement and estimated values of the parameters (asymptotic SE between brackets)

Lys	Variables to model Lys requirement ²	Latest added variable	R ² _{adj}	P ³	C	E	F	G	H
Total	5.1 -	-	91.0		.203 (.06)	9.9 (.7)	-	-	-
	5.2 age	Age	92.8	***	.435 (.08)	15.5 (1.1)	-.188 (.03)	-	-
	5.3 age, sex	Sex	92.9	NS	.432 (.08)	15.9 (1.1)	-.190 (.03)	-	-.58 (.4)
Digestible	5.1 -	-	88.0		.481 (.09)	8.8 (.3)	-	-	-
	5.2 age	Age	88.9	***	.506 (.09)	11.8 (1.0)	-.104 (.03)	-	-
	5.3 age, sex	Sex	89.4	**	.524 (.09)	12.2 (1.0)	-.102 (.03)	-	-.83 (.4)

1 $Breast = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + G^*year + H^*sex])))$, where: Breast = breast meat yield (g/g carcass); A = asymptotic breast meat yield; C = shape of the dose-response curve; Lys = dietary Lys level (g/kg); [...] = Lys 'requirement' (dietary Lys level at which Breast = 0.98*A), as a function of broiler age (d), year (e.g. 2002), and sex (1= males; 0 = females; 0.5 = as hatched).

2 Variables to model Lys requirement were added to the model for breast meat yield (see footnote 1) one-by-one.

Model 5.1: $Breast = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E])))$, basis model; for meaning of the variables and parameters, see footnote 1.

Model 5.2: $Breast = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age])))$; for meaning of the variables and parameters, see footnote 1.

Model 5.3: $Breast = 0.98^*A + 0.02^*A*(1 - \exp(-C^*(Lys - [E + F^*age + H^*sex])))$; for meaning of the variables and parameters, see footnote 1.

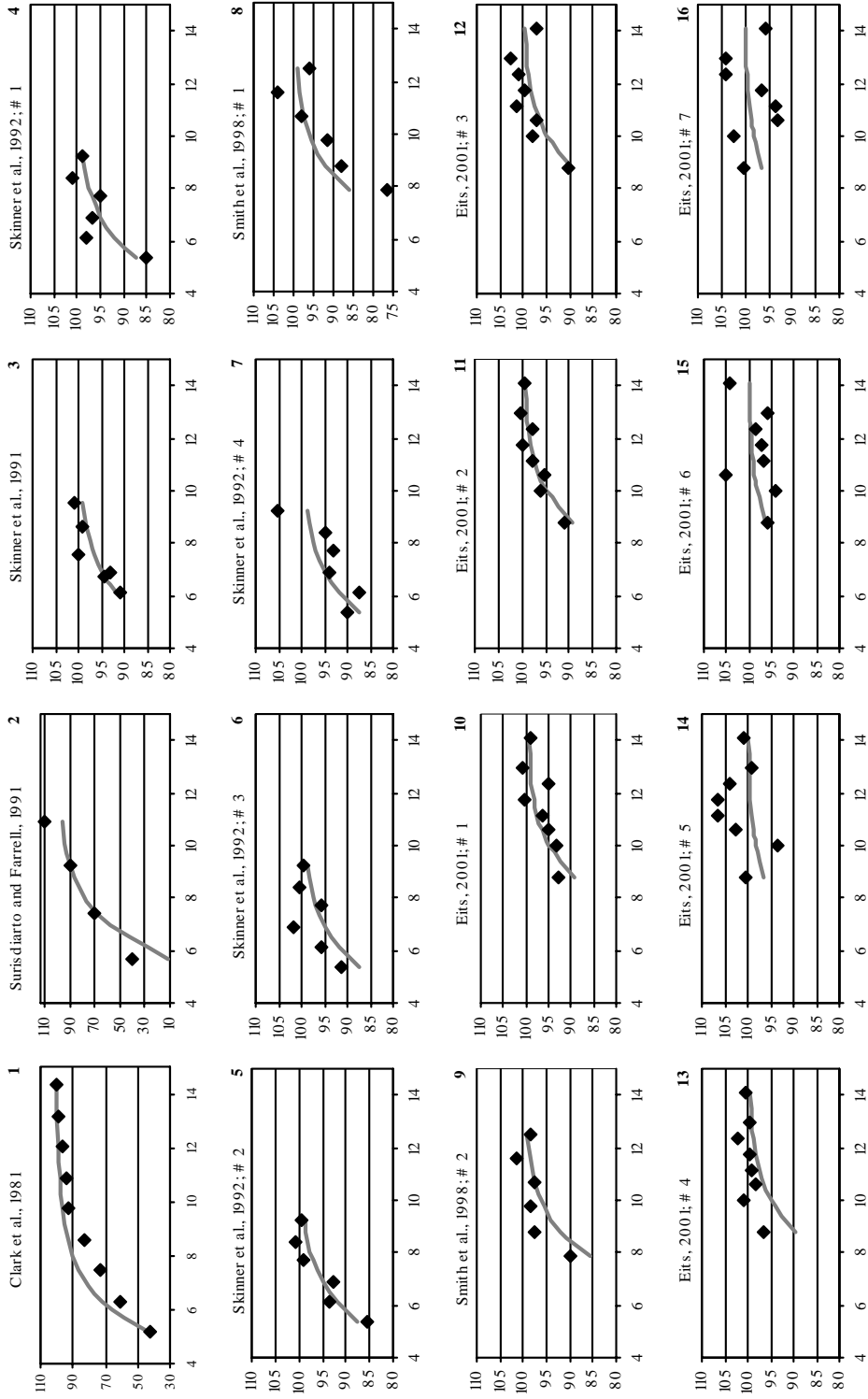
3 Significance of the contribution to the model by the latest added variable, NS: $P > 0.10$; * $0.10 > P > 0.05$; ** $0.05 > P > 0.01$; *** $P < 0.01$.

4 For the model on digestible Lys, a selection of 18 out of the 22 data sets in total was used (see text).

Estimated parameter values for the growth rate models are given in Table 2. Parameter *C* varied considerably among the different models, for example between 0.316 and 0.610 for total Lys (Table 2). In the basis model, with Lys and *A* as the only variables, parameter *E* represents an estimation of the balanced protein requirement over all data sets (11.2 and 9.14 g Lys/kg for total and digestible Lys, respectively). Parameter *F* is the estimated decrease in balanced protein requirement for each day increase in broiler age (for example, 0.0564 g Lys/kg diet/day for digestible Lys; Table 2). Parameter *G* corresponds to the effect of year on balanced protein requirement. The estimated value of 0.0935 (Table 2) suggests that by 10 years of genetic selection the Lys requirement for maximising growth rate increased with almost 1 g/kg.

Feed conversion ratio The models developed for FCR and the significance of the variables (age, year, sex) to predict Lys requirement for minimising FCR are presented in Table 3. The basis model, with total Lys and *A* as the only variables, explained 83.3% of the variation (Table 3). Age and year were the first and second most important variables to predict Lys requirement, and together ($R^2 = 97.0\%$) they explained 82% of the variance that was not explained by the basis model. Sex contributed significantly to the FCR model with digestible Lys ($P < 0.01$), but not to the FCR model with total Lys ($P > 0.10$; Table 3).

Estimated parameter values for the FCR models are given in Table 3. Parameter values can be compared (on a numerical basis) for the models for growth rate and FCR. Models that are compared are the ones that include all significant variables (Tables 2, 3). Parameters *C* in the models for FCR (0.540 and 0.544 for total and digestible Lys, respectively; Table 3) were lower than in the models for growth rate (0.610 and 0.759, for total and digestible Lys, respectively; Table 2). This means that FCR worsens relatively less than growth rate if Lys level becomes below the respective requirement. The overall balanced protein requirement (parameter *E* in the basis model) for FCR (11.9 and 10.7 g Lys/kg for total and digestible Lys, respectively; Table 3) was higher than for growth rate (11.2 and 9.14 g Lys/kg for total and digestible Lys, respectively; Table 2). The decrease in balanced protein requirement with age (parameter *F*) was slightly less for FCR (-0.0941 and -0.0467 g Lys/kg/d for total and digestible Lys, respectively; Table 3) than for growth rate (-0.0999 and -0.0564 g Lys/kg/d for total and digestible Lys, respectively; Table 2).



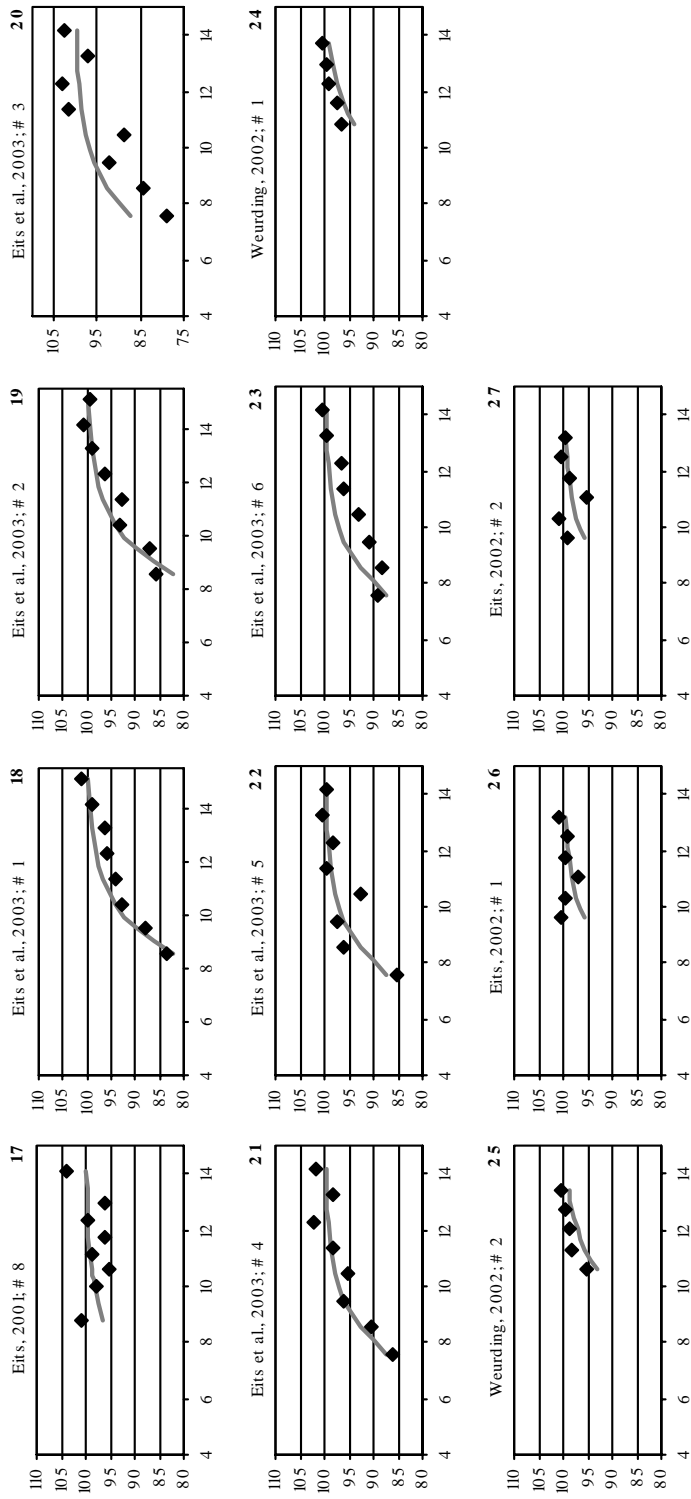
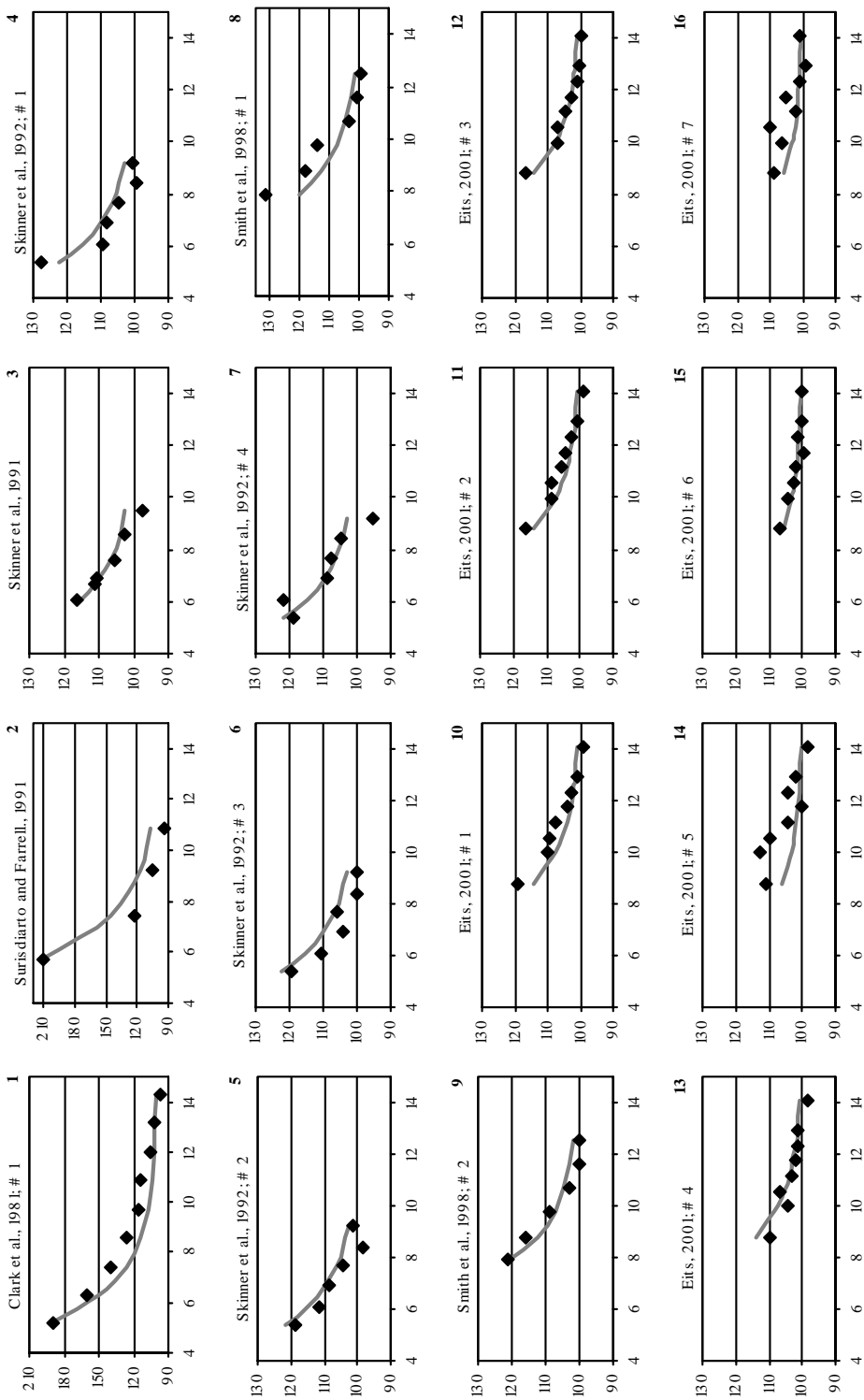


Figure 2. Growth rate (% of asymptotic growth rate in each individual trial) of broiler chickens at various dietary balanced protein levels (g total Lys/kg of diet). Experimental data (♦) and the response relationship (—) predicted by the general model based on total Lys level, asymptotic response, age and year. Data set number is represented by #.



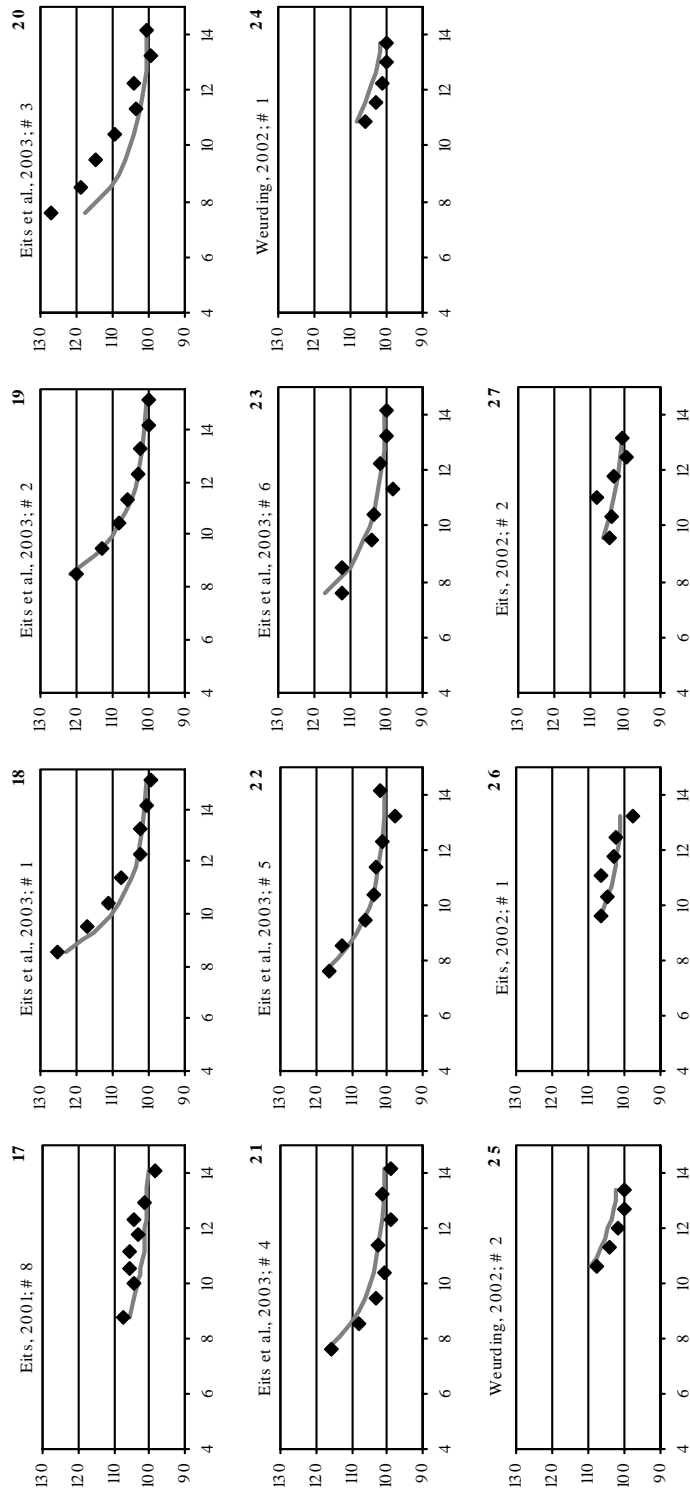
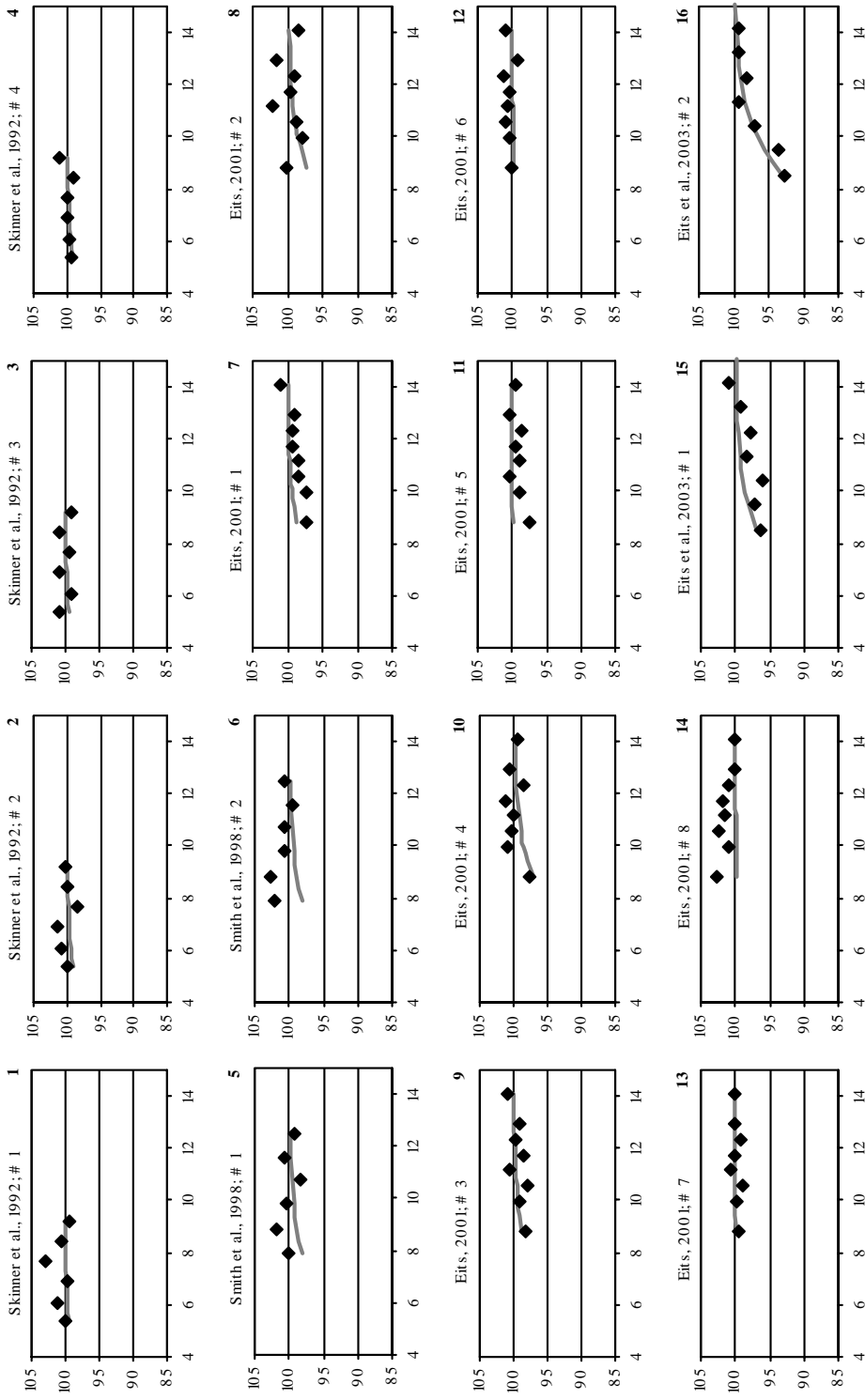


Figure 3. Feed conversion ratio (% of asymptotic feed conversion ratio in each individual trial) of broiler chickens at various dietary balanced protein levels (g total Lys/kg of diet). Experimental data (♦) and the response relationship (—) predicted by the general model based on total Lys level, asymptotic response, age and year. Data set number is represented by #.



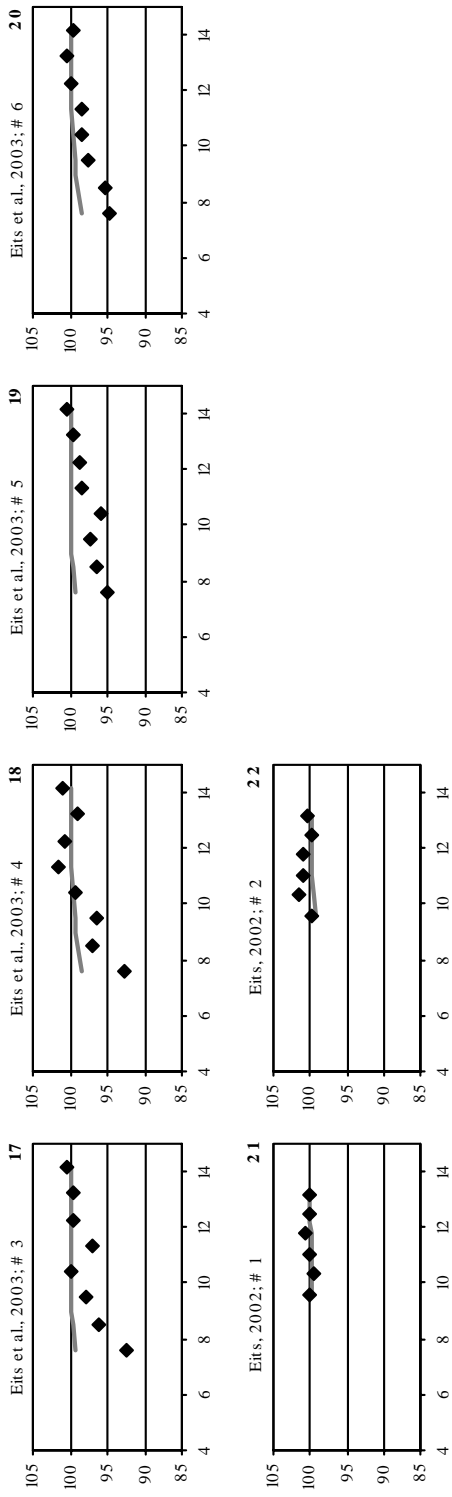
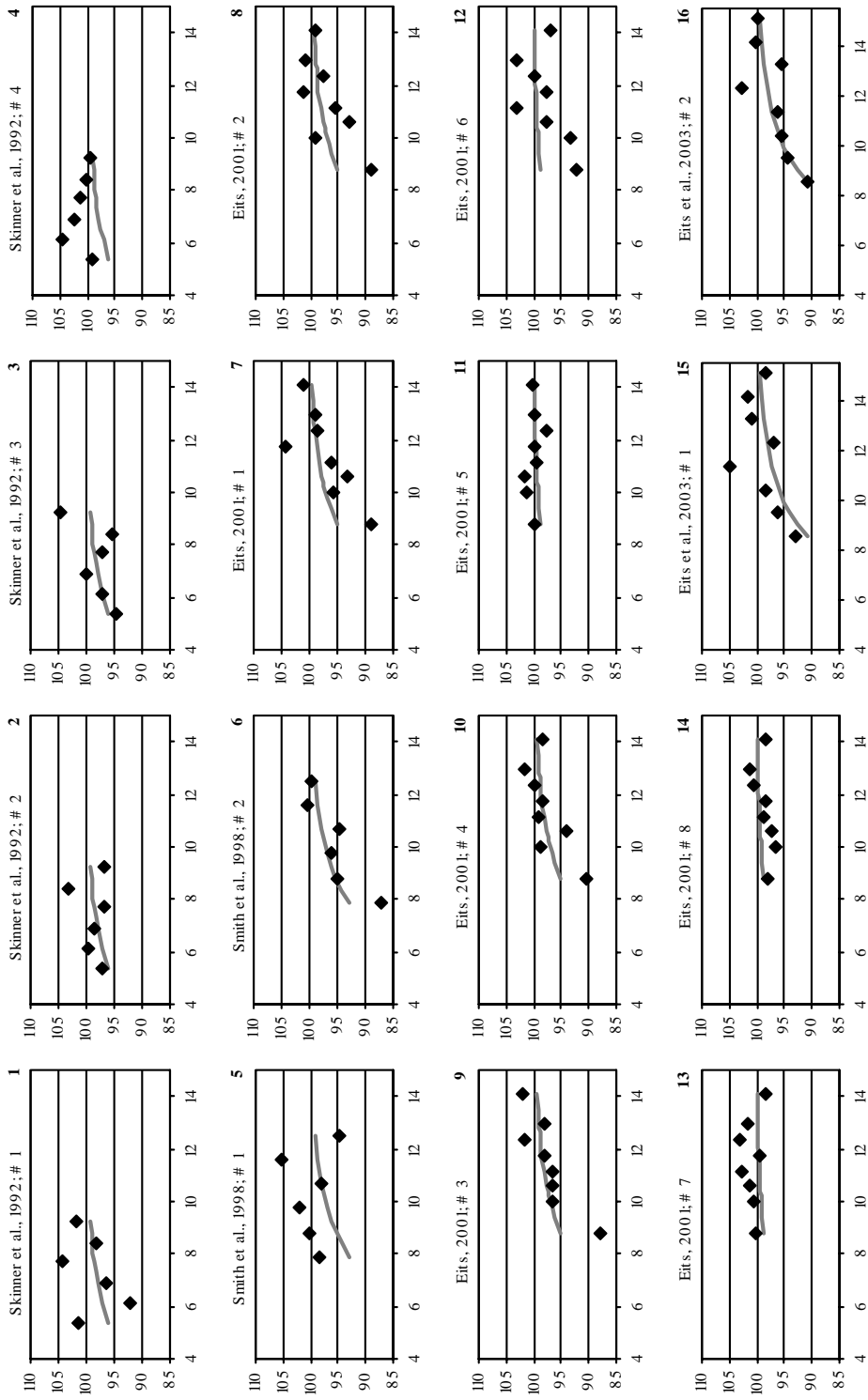


Figure 4. Carcass yield (% of asymptotic carcass yield in each individual trial) of broiler chickens at various dietary balanced protein levels (g total Lys/kg of diet). Experimental data (♦) and the response relationship (—) predicted by the general model based on total Lys level, asymptotic response, age and sex. Data set number is represented by #.



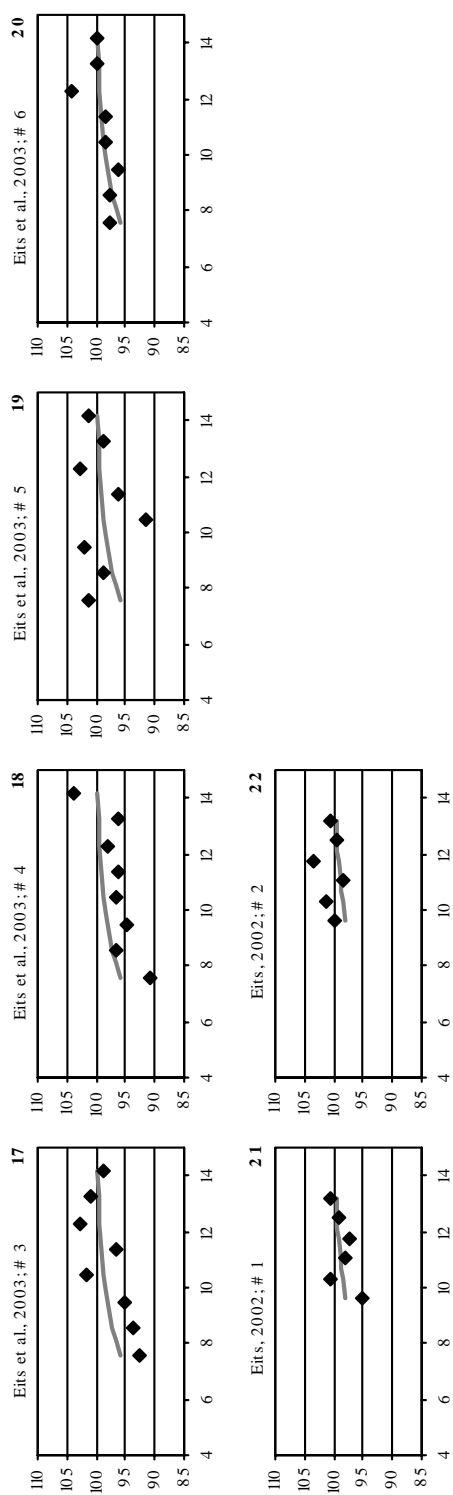


Figure 5. Breast meat yield (% of asymptotic breast meat yield in each individual trial) of broiler chickens at various dietary balanced protein levels (g total Lys/kg of diet). Experimental data (◆) and the response relationship (—) predicted by the general model based on total Lys level, asymptotic response and age. Data set number is represented by #.

Parameter G, the yearly increase in balanced protein requirement for FCR due to the genetic development (0.0572; Table 3) was only 61% of the effect on requirement for growth rate (0.0935; Table 2). The balanced protein requirement for FCR was higher for males than for females (parameter H), the magnitude of this difference being estimated as 0.716 g digestible Lys/kg (Table 3).

Carcass and breast meat yield The models developed for carcass and breast meat yield and the significance of the variables (age, sex) to predict Lys requirement for maximising carcass and breast meat yield are presented in Tables 4 and 5. Sex was not significant in the total Lys model for breast meat yield (Table 5).

Accuracy of the model

The experimental data and the dose response curves predicted by the model are presented in Figures 2-5. Data in Figures 2-5 were presented as relative responses (100% was defined as the asymptotic performance level in each individual data set) to enable a better comparison among data sets. The models used for the predictions in Figures 2-5 were based on total Lys and, for the variables age, year and sex, only the significant variables were included in the model (Tables 2-5).

Visual inspection of the graphs in Figures 2 (growth rate) and 3 (FCR) indicates that the dose response curves, using the same *C*-value for all data sets, give an acceptable to very good simulation of the experimental data for nearly all data sets. However, in panel 20 there is a bias between the model predictions and experimental data, for growth rate (Figure 2) as well as for FCR (Figure 3). This data set represents important compensatory responses of male broilers following a period with a low protein diet (Table 1). Figures 4 and 5 show that the effect of Lys on carcass and breast meat yield was less pronounced than on growth rate and FCR. The accuracy of the model predictions for carcass and breast meat yield was also less than for growth rate and FCR (Figures 2-5).

DISCUSSION

Results in relation to the objective

The objective was to develop a better method to model broiler performance as a function of dietary balanced protein level. Imposed restrictions were that the method should be applicable for modern commercial broiler

genotypes under field conditions, and that the method should be flexible and practical. Based on the following arguments, it can be concluded that the objective was realised. Firstly, the new method was based on recent studies with commercial broilers (Table 1). Secondly, the link to field conditions was realised by making field data on asymptotic performance level and on balanced protein requirement input for the model. Flexibility in type of broiler was obtained by the input variables age, year and sex. Finally, the new method remained practical by using only input data that are generally known by broiler nutritionists.

Results in Tables 2 and 3 together with Figures 2 and 3 showed that the new method gave an accurate description of growth rate and FCR for nearly all data sets that were used. It is note worthy that giving the shape of the response curve (C) the same value for all data sets turned out to be an acceptable simplification. The less accurate fit with compensatory responses of males fed a low protein starter diet (panel 20 in Figures 2 and 3) may reflect an underestimation of Lys requirement (see also panel b in Figure 1). Lys requirement may be increased in the period following a protein restriction (Chapter 3).

The accuracy of predictions in carcass and breast meat yield was lower than for growth rate and FCR (Figures 2-5). This may be explained by different definitions of carcass and breast meat among the experiments that were used for the model development. For example, the bias in the carcass yield predictions in some of the data sets (panels 17-20 in Figure 4) may be due to the fact that in that experiment only, carcasses were without skin and skin fat (Chapter 3). As another example, only in the study of Smith *et al.* (1998), carcasses included the abdominal fat pad. In this latter study, but not in the studies of our own laboratory, breast meat included skin and bone. Therefore, the accuracy in the predictions of carcass and breast meat yield responses may be improved if data sets with standardised carcass and breast meat data become available.

The ideal way of validating the complete model would be by means of independent data. However, this was not possible. The 27 data sets that satisfied the selection criteria were all needed to obtain a reliable selection of variables (age, year, sex) and estimation of parameter values (C , E , F , G and H). However, the Lys requirement part of the model (that is, the prediction from age, year and sex) can be validated by additional literature data. The effect of year (indicating genetic development) on Lys requirement is hardly or not investigated. The effects of age and sex on Lys requirement are better documented (NRC, 1994). In Figure 6, the Lys requirements for growth rate and FCR that were determined by the present

study are compared with the recommendations from NRC (1994). The relationships between total Lys requirement and age in the present study (Figure 6) were derived from the *D*- and *B*-parameter in model 3.3 for growth rate (Table 2) and in model 4.3 for FCR (Table 3). The estimated Lys requirement at a given age was up to 28% higher than according to NRC (1994), particularly for the first three weeks of age (Figure 6). Similar results were obtained in a study reported by CVB (1996), which is now confirmed by the present study based on 27 data sets.

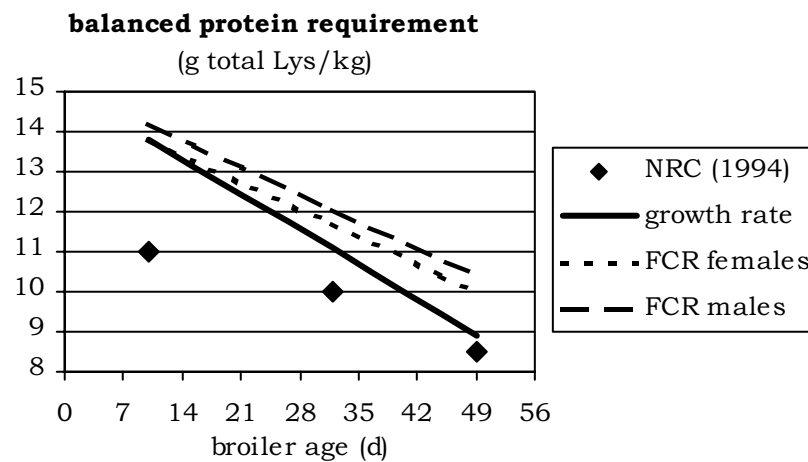


Figure 6. *Balanced protein requirement of broiler chickens as affected by age. Requirements derived from the present study (for optimising growth rate or feed conversion ratio (FCR)) and according to NRC (1994) (♦).*

Application of the new method

As an example, a nutritionist may want to determine the economic optimal balanced protein level in the diet for broilers of a given age, sex and breed. To do so, accurate dose-response curves in broiler performance to dietary balanced protein level are needed. The model developed in the present study can easily predict such dose-response curves. The nutritionist should give input values for the two additional model variables: asymptotic performance (*A*) and Lys requirement.

The *A* value and Lys requirement should represent the situation in the field, averaged over all the farms that will use the same broiler diet. The *A* value corresponds to the performance level of the broilers under study, using a dietary balanced protein level that is higher than the requirement. This *A* value is best estimated by the nutritionist himself, based on the

actual average broiler performance, in combination with experience about the sensitivity of broiler performance to increments in Lys level at practical Lys levels. This expert knowledge will in most cases deliver a more correct estimate for A than by any other means, for example a feeding trial, growth simulation model or generalized values from the breeding company.

Dietary energy interacts with dietary protein for the effect on broiler performance (Chapter 1). Dietary energy level was, however, not included as a variable in the developed models for growth rate and FCR. One reason was that the calculated energy levels varied little among the data sets used for the model development. Typical energy levels for broiler diets were applied in all these studies. Moreover, dietary energy levels in the various data sets were expressed on different scales, which makes comparison over data sets complicated. In principle, and as an alternative to using empirical data, theoretical arguments could be used to include dietary energy as a variable into the models. For example, the concept that poultry tend to eat to meet their energy needs (NRC, 1994). This concept implies that the effect of decreasing energy level could be accounted for in the model for growth rate by a proportional reduction in Lys requirement. However, this concept appears to be invalid for imbalanced diets. In Figure 7, it is shown that particularly at very low Lys to energy ratios, feed intake of broilers is not only affected by energy level but by Lys level as well. The response shown in Figure 7 is in accordance with the theory stated by NRC (1994) on the effect of amino acids level on *ad libitum* feed intake. Thus, a good way of including energy level as a variable in the models of the present study is not available yet. Therefore bias may occur with the application of the models if the energy level of a diet substantially differs from the average energy level in the data sets used for the present study. The model predictions are most reliable for diets with typical energy levels, for example 13.4 MJ MEn/kg (NRC, 1994) or 12.6 MJ MEn-broilers/kg (CVB, 1996) for a broiler grower diet.

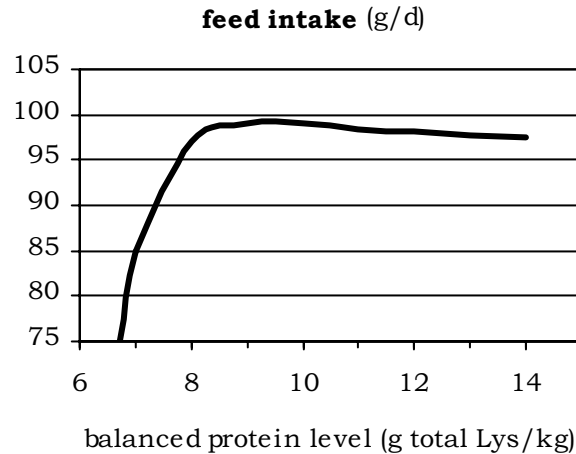


Figure 7. Feed intake as affected by dietary balanced protein level. Feed intake calculated as growth rate (model 3.3; Table 2) multiplied by FCR (model 4.3; Table 3). Input values used were 65 g/d (A; growth rate), 1.50 (A; FCR), 21 d (age) and 2003 (year).

Lys requirement may be predicted from age, year and sex by means of a formula that was developed in the present study. Predictions by this formula are supposed to be fairly accurate for modern broilers, as they are based on many recent data sets. However, if a nutritionist has data on Lys requirement under his field conditions that are more accurate than the predictions by the formula, then he can use his own estimate for Lys requirement to predict the dose-response curves.

Added value of the new method

Dose-response relationships to dietary balanced protein, needed for formulation of optimal broiler diets, may be quantified in a feeding experiment (for example, Clark *et al.*, 1982; Smith *et al.*, 1998). Many interacting factors are included in the results of such *in vivo* tests. Due to its empirical nature, however, this approach has considerable drawbacks. A lot of unexplained variation exists between dose response relationships from different experiments. Therefore, the relationships may be only valid under the exact conditions (in terms of bird characteristics, feed and environment) of the specific experiment. The practical consequence is that, following any change in conditions in a specific broiler operation, new expensive and time-consuming experiments will be required to validate the dose responses under the new conditions. The method developed in the present study is based on 27 data sets. Moreover, additional variables were included that account for much of the quantitative variation in dose-

responses to dietary balanced protein between data sets. Therefore, dose-responses predicted by the new method should be more flexible and accurate under a wider range of conditions than predictions derived from one or a few feeding trials.

Alternatively, dose response relationships may be predicted by a mechanistic growth simulation model (Gous, 1998). For example the model described by Emmans (1981) that includes a complete theory on feed intake regulation and nutrient partitioning in the body of broiler chickens. Scientifically, such a model is of importance to identify the most relevant gaps in existing knowledge. Broiler nutritionists, however, are usually not familiar with some of the required input data (for example, minimal fat to protein ratio in the body; Emmans, 1981). Moreover, most broiler nutritionists do not have the time to study the underlying biological theories, which is essential to understand the model predictions. The model developed in the present study only includes variables of which experienced nutritionists know the actual value for their practical conditions. Besides, the model is straightforward and thus relatively easy to understand.

ACKNOWLEDGMENTS

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Chapter 5

**Dietary Balanced Protein in Broiler Chickens: an
Economic Analysis**

Paper by: R. M. Eits, G. W. J. Giesen, R. P. Kwakkel, M. W. A. Verstegen
and L. A. den Hartog. Submitted to British Poultry Science.

ABSTRACT An economic model was developed that calculates economic optimal dietary balanced protein levels (DBP) for broiler chickens, based on technical input and prices of meat and feed. Technical input on broiler responses to DBP (growth rate, feed conversion, carcass yield and breast meat yield) was obtained from the model described in Chapter 4.

Changes in broiler age, price of protein-rich raw materials and large changes (40%) in meat prices result in economic relevant differences in DBP for maximum profit. Effects of changes in sex or feed price on DBP for maximum profit are negligible. Formulating diets for maximum profit instead of maximum broiler performance can strongly increase profitability of a broiler production enterprise. DBP for maximum profitability depend on how the broilers are marketed; as whole birds, carcass or cut up.

INTRODUCTION

Dietary protein level in broiler diets strongly affects growth rate and meat yield (Chapter 4) as well as feed cost and, thereby, profitability of a broiler production enterprise. NRC (1994) therefore stated that it would be desirable to have mathematical models available that would facilitate the selection of the most economic nutrient levels. A technical as well as an economic model is required for this aim. A technical model that predicts broiler performance and meat yield responses to dietary balanced protein has been described in Chapter 4. Just a few economic models were published that evaluate the economics of broiler nutrition, for example by Hurwitz *et al.* (1985), Gonzalez *et al.* (1992), Pack and Schutte (1995) and Fisher and Gous (2000). However, these economic models as such were inappropriate for economic evaluation of broiler responses to dietary balanced protein level over a constant age period, as in the technical model in Chapter 4.

Therefore, the first objective of this study was to develop an economic model to determine economic optimal protein levels from price data and performance data (from the technical model). The second objective was to study the effect of broiler age and sex and of prices of feed and meat on the economic optimal protein level.

MODEL DESCRIPTION

Concept of the model

An economic model was developed that calculates the effect of protein level in the diet on feed costs, revenues and thereby on 'returns over feed cost' per broiler. Returns over feed cost were defined as gross margin.

It is common in practise that nutritional modifications do not affect the slaughtering age. Therefore, analyses in this study were made over a constant age period. Consequently, it is reasonable to assume that feed costs are the only costs affected by protein level in the diet. Slaughtering costs per broiler, for example, are more or less constant within the range of broiler weights that a given slaughterhouse can handle.

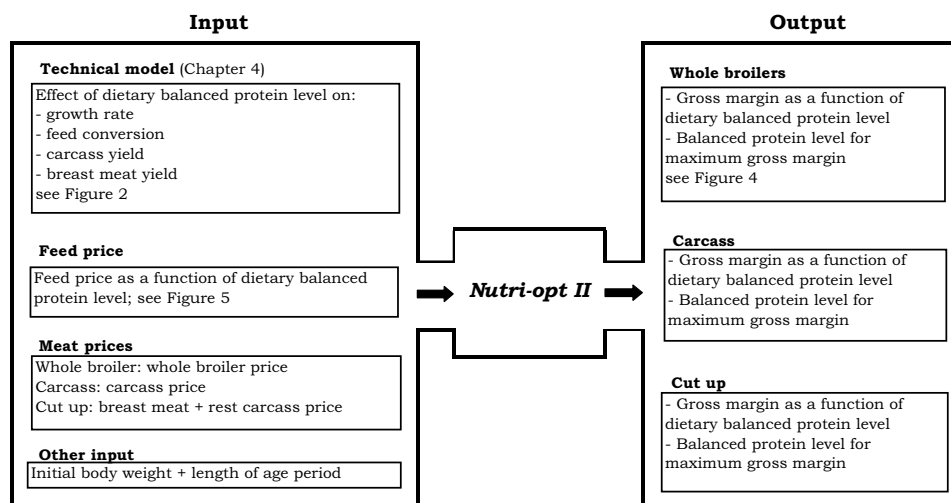


Figure 1. Overview of input and output for the economic model “Nutri-opt II”.

Revenues depend on how the broilers are marketed: as whole broilers, as whole carcass (slaughtered broiler) or as cut up meat parts. Cut up broilers were defined here as consisting of two components: breast meat and the remainder (rest carcass).

Calculations of the economic model apply to one age period, for example the age period during which the grower or finisher diet is fed. It was assumed that each gram difference in broiler weight (or weight of carcass, breast meat, feed intake, etc.) at the end of an age period results in the same difference at slaughter age.

Dietary protein was defined as protein balanced for amino acids (balanced protein) with Lys as reference amino acid and the other essential amino acids kept in constant and balanced proportions relative to Lys. Lys was expressed on a digestible basis. Dietary energy and other nutrients were assumed to be at constant and adequate levels, for example meeting NRC (1994) specifications. An overview of input and output for the economic model is presented in Figure 1.

An Excel spreadsheet program, called Nutri-opt II, was developed to facilitate the calculations of Lys level on technical performance (equations [1]-[4]) and economical performance (equations [5]-[10]). Nutri-opt II runs the calculations of gross margin for Lys levels increasing from 6.0 up to 13.0 g/kg in steps of 0.1 g/kg (70 Lys levels in total).

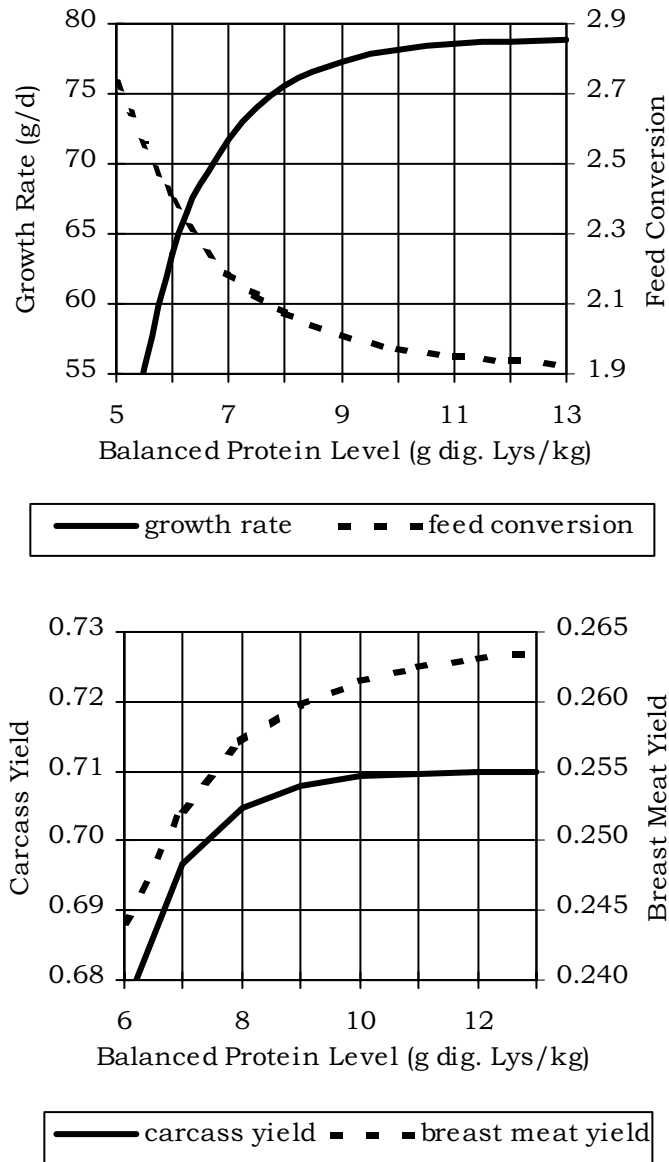


Figure 2. Broiler responses to dietary balanced protein level. Reference situation. Carcass yield (g/g body weight) and breast meat yield (g/g carcass).

Model input

Technical input Models were developed to simulate growth rate (g/d), feed conversion, carcass yield (g/g live weight) and breast meat yield (g/g carcass) of broilers as a response to dietary balanced protein level (Lys as a reference) (Chapter 4). From Chapter 4, those equations were selected that include significant variables only and in which Lys was expressed on a digestible basis:

$$\text{Growth rate} = 0.98 * A + 0.02 * A * (1 - e^{-0.759 * (Lys - [10.8 - 0.0564 * age])}) \quad [1]$$

$$\text{Feed conversion} = 1.02 * A - 0.02 * A * (1 - e^{-0.544 * (Lys - [11.7 - 0.0467 * age + 0.716 * sex])}) \quad [2]$$

$$\text{Carcass yield} = 0.98 * A + 0.02 * A * (1 - e^{-0.947 * (Lys - [9.5 - 0.061 * age - 1.24 * sex])}) \quad [3]$$

$$\text{Breast meat yield} = 0.98 * A + 0.02 * A * (1 - e^{-0.524 * (Lys - [12.2 - 0.102 * age - 0.83 * sex])}) \quad [4]$$

where: *A* is asymptotic performance level (for example, *A* is growth rate at a Lys level that is higher than the Lys requirement for maximising growth rate), Lys is digestible Lys level in the diet (g/kg) and age is average broiler age for the relevant age period (d). Sex is expressed as 0 for females, 1 for males and 0.5 for as hatched. Figure 2 gives an illustration of each of the four broiler performance models (equations [1] to [4]). It was assumed that nutritional responses during an age period are independent of previous nutrition.

Prices and other input Feed price (€/100 kg) was defined as cost of the raw materials. Several years of experience in Nutreco companies showed that the relationship between feed price and dietary balanced protein level can accurately be described by a quadratic model:

$$\text{Feed price} = a * Lys^2 + b * Lys + c \quad [5]$$

where: Lys is dietary Lys level (g/kg), which is used as the indicator of dietary balanced protein level. *A*, *b* and *c* are regression coefficients that depend on the prices and quality of the raw materials in the feed.

Prices of live broilers, carcass, breast meat and rest carcass were in €/kg. The other input was broiler weight at the start of the relevant age period (initial weight; kg) and length of the age period (days).

Model output

The economic model calculates the effect of dietary balanced protein level (Lys as a reference) on revenues, feed costs and, thereby, on gross margin (Figure 3). From these data, the economic optimal Lys level (Lys level for maximum gross margin; Lys_{econ}) can be determined (Figure 3).

The effect of Lys on revenues (€/broiler) is calculated from the effect that Lys has on broiler weight, carcass yield and breast meat yield:

$$Revenues_{\text{whole broiler}} = BW * \text{whole broiler price} \quad [6]$$

$$Revenues_{\text{carcass}} = BW * \text{carcass yield} * \text{carcass price} \quad [7]$$

$$Revenues_{\text{cut up}} = BW * \text{carcass yield} * [\text{breast meat yield} * \text{breast meat price} + (1 - \text{breast meat yield}) * \text{rest carcass price}] \quad [8]$$

where: carcass yield and breast meat yield are predicted using the equations [3] and [4]. BW is broiler weight at the end of the age period (kg). The effect of Lys on BW is calculated from the effect that Lys has on growth rate:

$$BW = IBW + \text{growth rate}/1000 * \text{days} \quad [9]$$

where: growth rate is predicted using equation [1]. IBW is initial broiler weight (kg) and days refer to the length of the age period (d).

The effect of Lys on feed cost (€/broiler) is calculated from the effect that Lys has on growth rate, feed conversion and feed price:

$$Feed\ cost = \text{growth rate}/1000 * \text{days} * \text{feed conversion} * \text{feed price}/100 \quad [10]$$

where: growth rate, feed conversion and feed price are predicted using the equations [1], [2] and [5]. Gross margin (€/broiler) is calculated as the difference between revenues and feed cost.

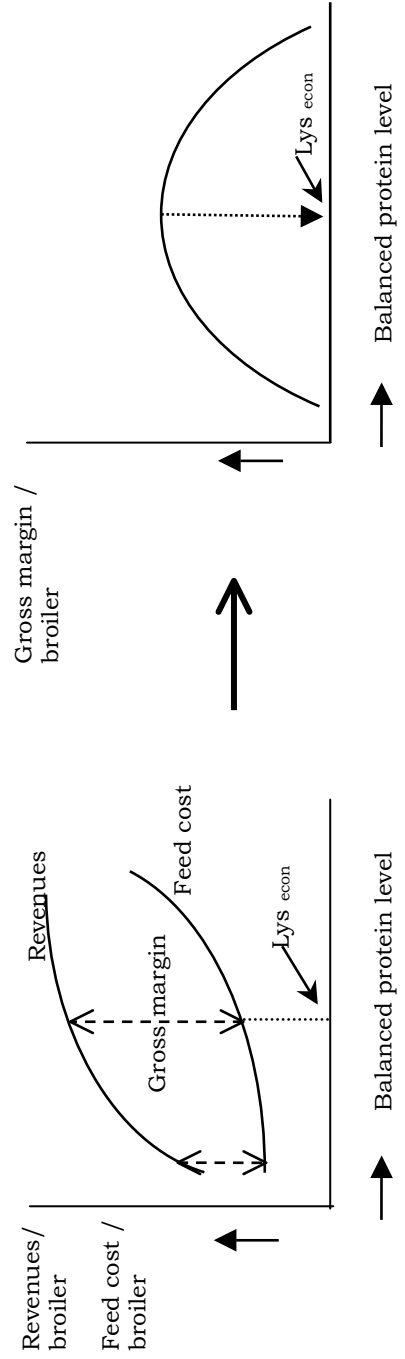


Figure 3. The effect of balanced protein level in a broiler diet on revenues, feed cost and, thereby, on gross margin per broiler. Lys_{econ} is balanced protein level for maximum gross margin.

Table 1. Asymptotic performance level ¹ in the reference situation and in alternative situations with different ages or sex

	Reference	Alternative: lower age	Alternative: higher age	Alternative: sex = male	Alternative: sex = female
Age (d)	21-42	7-28	35-56	21-42	21-42
Sex	as hatched	as hatched	as hatched	male	female
Growth rate (g/d)	78.9	55.4	76.1	87.7	70.1
Feed conversion	1.92	1.52	2.37	1.87	1.98
Carcass yield (g/g body weight)	0.710	0.686	0.725	0.716	0.704
Breast meat yield (g/g carcass)	0.264	0.251	0.276	0.263	0.266
Initial body weight (kg)	0.824	0.165	1.902	0.851	0.797

¹ Asymptotic performance level: variable A in equations [1] to [4] (see text); data derived from Hybro (2003).

Table 2. Effect of broiler age and sex on balanced protein level for maximum gross margin (Lys_{econ}) and the reduction in gross margin (Δ gross margin) if balanced protein level is not adapted to broiler age or sex.

	WHOLE BROILER ¹			CARCASS ¹		CUT UP ¹	
	Lys_{econ} (g/kg)	Δ gross margin ² (€-cents/broiler)	Lys_{econ} ² (g/kg)	Δ gross margin ² (€-cents/broiler)	Lys_{econ} (g/kg)	Δ gross margin ² (€-cents/broiler)	
Age							
	9.6	0.38	10.2	0.56	10.6	0.63	
7-28 d							
	8.9	-	9.4	-	9.8	-	
21-42 d (reference)							
	8.2	0.50	8.8	0.47	9.2	0.52	
35-56 d							
Sex							
	8.7	0.04	9.5	0.00	9.9	0.02	
female							
	8.9	-	9.4	-	9.8	-	
as hatched (reference)							
	9.0	0.02	9.5	0.00	9.8	0.00	
male							

¹ Results are presented separately depending on how broilers are marketed: as whole broilers, as carcass (slaughtered broilers) or as cut up meat parts;

² Reduction in gross margin if balanced protein level is not changed to Lys_{econ} for the alternative age or sex, but maintained at Lys_{econ} for the reference situation.

MODEL SIMULATIONS

Calculations

Simulations were done for a reference situation (as defined below) as well as for situations with alternative age, sex, meat prices or feed prices. For all situations, the Lys level for maximum gross margin (Lys_{econ}) was determined (Figure 3). In addition, it was calculated how much gross margin would be reduced (Δ gross margin) if, in the alternative situation, not the optimal Lys level (Lys_{econ}) is fed, but the Lys level that is optimal in the reference situation. The calculation of Δ gross margin can be explained from Figure 4. First, gross margin as a function of Lys level is calculated both for the reference situation and for the alternative situation (for example, a higher broiler price). L1 is Lys_{econ} for the reference situation, whereas L2 is Lys_{econ} for the alternative situation (Figure 4). The Δ gross margin if Lys is not adapted to the higher broiler price (L2 instead of L1) is the difference in gross margin (at the alternative situation) at $Lys = L2$ (GM2) and at $Lys = L1$ (GM1).

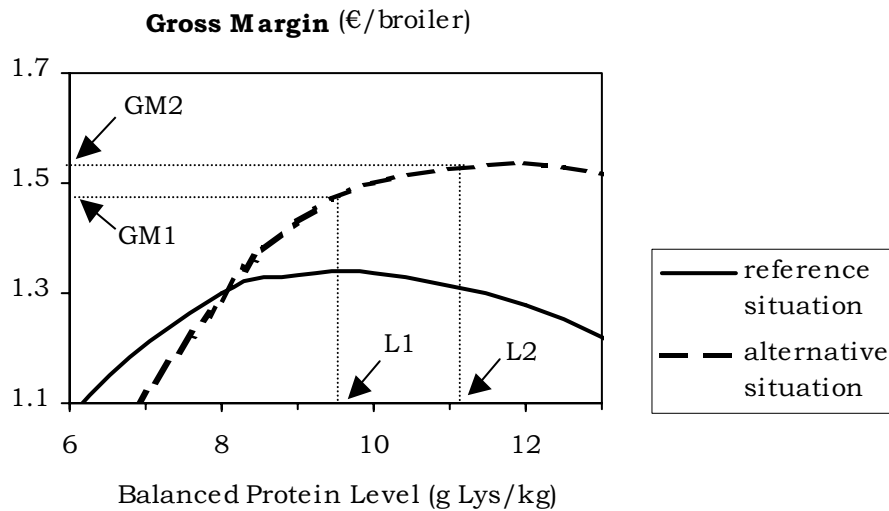


Figure 4. Example of the calculation of extra profit ($GM2-GM1$) that results from adapting the balanced protein level from the level that is optimal for the reference situation ($L1$) to the level that is optimal for the new scenario with increased broiler price ($L2$). $GM1$ and $GM2$ refer to gross margin at the new scenario, at $L1$ and $L2$, respectively.

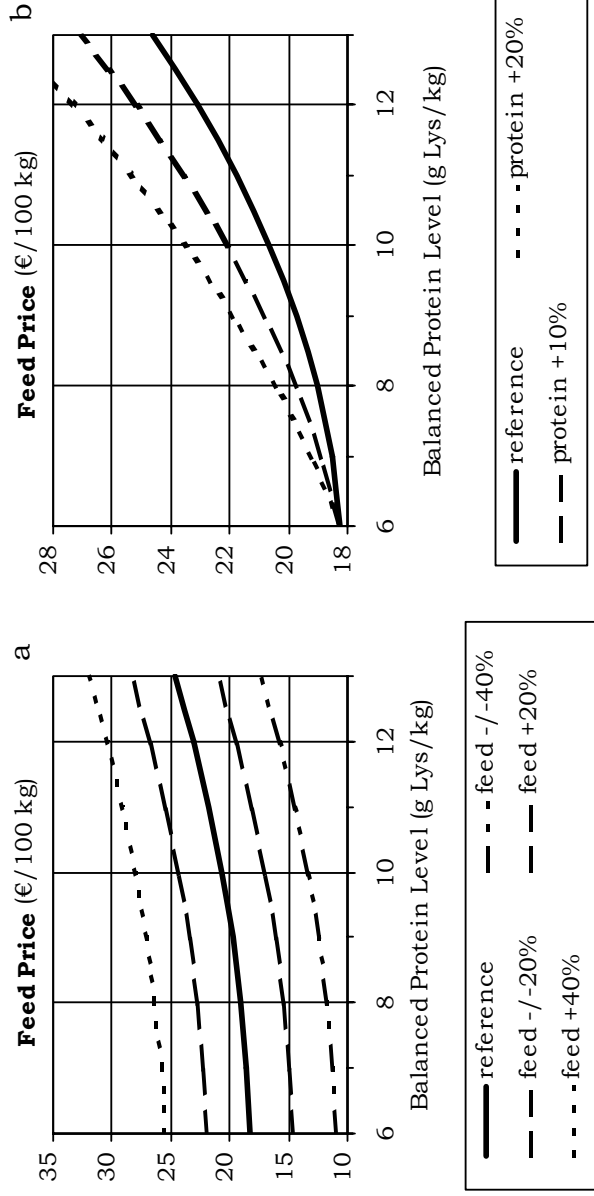


Figure 5. Feed price as a function of dietary balanced protein level in the reference situation and in alternative feed price situations. Variation in feed price level (figure 5a) and variation in price of protein-rich raw materials (figure 5b) Variation in feed price was initiated by an increase or decrease of the feed price at all protein levels by 20 or 40% of the feed price at Lys = 6.0 g/kg. Variation in protein-rich raw materials was initiated by a protein level dependent increase in feed price, varying from no increase at 6.0 g/kg Lys up to 10 or 20% increase at 13.0 g/kg Lys. Reductions in price of protein-rich raw materials were not evaluated as it resulted in a constant or decreasing feed price for Lys levels between 6 and 8 g/kg, which is unlikely to happen in practise.

The reference situation was defined as as hatched broilers of 21 to 42 d. The related performance variables are presented in Table 1 and the dose-response relationships for the reference situation are in Figure 2. Prices of meat and feed for the reference situation are given in Table 3 and Figure 5, respectively, and were based on actual prices in The Netherlands (December, 2003).

Table 3. Effect of meat prices on Lys level for maximum gross margin (Lys_{Secon}) and the reduction in gross margin (Δ gross margin) if Lys level is not adapted to the alternative meat price ¹

Price of	Relative to reference price	Actual price (€/kg)	Lys_{Secon} (g/kg)	Δ gross margin ¹ (€-cents/broiler)
Whole broiler	- 40%	0.42	8.4	0.23
	- 20%	0.56	8.7	0.06
	reference	0.70	8.9	-
	+ 20%	0.84	9.0	0.01
	+ 40%	0.98	9.2	0.08
Carcass	- 40%	0.90	9.0	0.19
	- 20%	1.20	9.2	0.03
	reference	1.50	9.4	-
	+ 20%	1.80	9.6	0.05
	+ 40%	2.10	9.7	0.17
Cut up	- 40%	2.10 / 0.78 ²	9.4	0.22
	- 20%	2.80 / 1.04 ²	9.6	0.04
	reference	3.50 / 1.30 ²	9.8	-
	+ 20%	4.20 / 1.56 ²	10.0	0.07
	+ 40%	4.90 / 1.82 ²	10.2	0.22

¹ Reduction in gross margin if Lys level is not changed to Lys_{Secon} for the alternative meat price, but maintained at Lys_{Secon} for the reference meat price;

² Prices of breast meat and rest carcass, respectively.

Details of the alternative age and sex situations are given in Table 1. In the alternative meat price situations, meat prices were 20 or 40% higher or lower than in the reference situation (Table 3). Two alternative feed price scenarios were studied: variation in feed price level (Figure 5a) and variation in price of the protein-rich raw materials, for example soy bean meal and synthetic amino acids (Figure 5b).

Results

In the reference situation, Lys levels for maximum gross margin (Lys_{econ}) are 8.9 g/kg for whole broilers, 9.4 g/kg for carcass and 9.8 g/kg for cut up.

Feeding broilers that will be sold as cut up with the optimal grower diet (21-42 d) for whole broilers ($Lys = 8.9$ g/kg) instead of the optimal grower diet for cut up ($Lys = 9.8$ g/kg) would decrease gross margin over the grower period with 1.45 €-cents per broiler. Similarly, feeding broilers that will be sold as whole broilers with the optimal grower diet for cut up ($Lys = 9.8$ g/kg) instead of the optimal grower diet for whole broilers ($Lys = 8.9$ g/kg) would decrease gross margin over the grower period with 0.85 €-cents per broiler. These data show that economic relevant differences exist in Lys_{econ} for broilers marketed as whole broilers, carcass or cut up, respectively.

Table 2 shows the effect of broiler age on Lys_{econ} , and the reduction in gross margin (Δ gross margin) if Lys level is not adapted to broiler age. For young broilers (age = 7-28 d), Lys_{econ} was higher than for the reference situation (age = 21-42 d). For example for 'whole broilers', Lys_{econ} for young broilers was 9.6 g/kg compared to 8.9 g/kg for the reference situation (Table 2). If, instead of the optimal diet ($Lys = 9.6$ g/kg), the reference diet ($Lys = 8.9$ g/kg) is used for young broilers, gross margin would be reduced by 0.38 €-cents/broiler (Δ gross margin for 'whole broilers'; Table 2). For carcass and cut-up, Δ gross margins were 0.56 and 0.63 €-cents/broiler, respectively. For older broilers (age = 35-56 d), Lys_{econ} was lower than for the reference situation and Δ gross margins were between 0.47 and 0.52 €-cents/broiler (Table 2).

The differences in Lys_{econ} for males and females were 0.3 g/kg or less (Table 2). Consequently, the effects of sex on Δ gross margin were negligible. The effects of age on Lys_{econ} were 5 to 10 times the effects of sex on Lys_{econ} . Effects of age and sex on Lys_{econ} and Δ gross margin were independent of how the broilers are marketed (Table 2).

The effects of meat prices on Lys_{econ} and Δ gross margin are given in Table 3. A 20% increase or decrease in meat prices resulted in minor effects on Lys_{econ} ; Lys_{econ} levels differed 0.1 or 0.2 g/kg from the Lys_{econ} levels at the reference situation (Table 3). As a result, effects on Δ gross margin were only 0.07 €-cents/broiler or less. The effect of a 40% increase (or decrease) in meat prices on Δ gross margin was more than three times the effect of a 20% increase (or decrease) in meat prices (Table 3). In other words: although meat price affects Lys_{econ} about proportionally, meat price affects Δ gross margin more than proportionally. This disproportionality is a consequence of the 'parabolic' relationship between gross margin and Lys level (Figure 3).

Feed price level had negligible effects on Lys_{econ} and Δ gross margin (Table 4). That feed price level has negligible effects on Lys_{econ} can be explained from Figure 3. Lys_{econ} is the Lys level at which the difference between revenues and feed cost is maximal (Figure 3). Feed cost is calculated as feed intake multiplied by feed price. Feed intake, which is calculated from growth rate and feed conversion, is about constant in the range of Lys levels in which Lys_{econ} is always determined in the present study (between 8 and 11 g/kg) (Chapter 4). Therefore, in this range of Lys levels where feed intake is about constant, an increase in feed price increases feed cost with a fixed amount, independent of Lys level. Consequently, in this range of Lys levels, the Lys level at which the difference between revenues and feed costs is maximal ($=Lys_{econ}$) is not affected by feed price level.

An increase in price of protein-rich raw materials decreased Lys_{econ} strongly and about proportionally (Table 4). The effect was independent of how the broilers are marketed (whole broilers, carcass or cut up). This Lys level dependent increase in feed price (Figure 5b) results in a Lys level dependent increase in feed cost. Revenues are not affected by price of protein-rich raw materials. Consequently, the Lys level at which the difference between revenues and feed cost is maximal ($=Lys_{econ}$) becomes lower than for the reference situation.

In summary, the model calculations revealed that broiler age and price of protein-rich raw materials (+ 20%) have the largest effects on Lys_{econ} (> 0.6 g/kg) and on Δ gross margin (> 0.38 €-cents/broiler). A large change (40%) in meat price and a 10% price increase of protein-rich raw materials have only small effects on Lys_{econ} (> 0.3 g/kg) and on Δ gross margin. Effects of sex, feed price level and small changes in meat price (20%) on Lys_{econ} and Δ gross margin were negligible.

Table 4. Effect of feed price level and of prices of protein-rich raw materials on balanced protein level for maximum gross margin (Lys_{econ}) and the reduction in gross margin (Δ gross margin) if the balanced protein level is not adapted to the alternative feed price scenario.

	WHOLE BROILER ¹			CARCASS ¹		CUT UP ¹	
	Lys_{econ} (g/kg)	Δ gross margin ² (€-cents/broiler)	Lys_{econ} (g/kg)	Δ gross margin ² (€-cents/broiler)	Lys_{econ} (g/kg)	Δ gross margin ² (€-cents/broiler)	
Feed price level ³	-/- 40%	8.8	0.03	9.4	0.00	9.8	0.00
	-/- 20%	8.8	0.01	9.4	0.00	9.8	0.00
	reference	8.9	-	9.4	-	9.8	-
	+ 20%	8.9	0.00	9.5	0.01	9.9	0.01
	+ 40%	9.0	0.00	9.5	0.01	9.9	0.02
Price of protein-rich raw materials ³	reference	8.9	-	9.4	-	9.8	-
	+ 10%	8.4	0.34	9.1	0.18	9.5	0.16
	+ 20%	8.0	1.16	8.7	0.75	9.2	0.70

¹ Results are presented separately depending on how broilers are marketed: as whole broilers, as carcass (slaughtered broilers) or as cut up meat parts;

² Reduction in gross margin if balanced protein level is not changed to Lys_{econ} for the alternative feed price scenario, but maintained at Lys_{econ} for the reference situation. Feed price (€/100kg) in the reference situation = $0.1034 * Lys^2 - 1.0513 * Lys + 20.83$;

³ For description of situations with alternative feed price level or alternative price of protein-rich raw materials: see Figure 5.

DISCUSSION

The economic model

Most research in broiler nutrition concentrates on maximising broiler performance while economic factors are often ignored. The strength of the model developed in the present study is that it enables calculation of the economic optimal protein levels. This can help nutritionists in formulating diets for maximum profit or, at least, enables calculation of the extra investment that is made by formulating diets for maximum broiler performance instead of maximum profit (see below). Another strength is that the model considers how broilers are marketed; as whole broilers, carcass or cut up. The model has a weak point in that it assumes that each gram difference in broiler weight (or weight of carcass, breast meat, feed intake, etc.) at the end of an age period results in the same difference at slaughter age. More data are needed to investigate the validity of this assumption. It should be noted, however, that not making this assumption would make the model much more complex as the model should then consider the issue of compensatory performance. Another weak point of the model may be that it does not consider the (environmental) cost of nitrogen excretion. The relevancy of this subject depends on local conditions, for example density of animal production or local regulations. The present model was developed for more general application and did therefore not consider this subject.

Just a few other models were published that evaluate the economics of broiler nutrition, for example by Hurwitz *et al.* (1985), Gonzalez *et al.* (1992), Pack and Schutte (1995) and Fisher and Gous (2000). Pack and Schutte (1995) developed a model to calculate economic optimal Met + Cys levels from broiler response data in feed conversion and breast meat yield. Pack and Schutte (1995) concluded that for selling cut up broilers, higher Met + Cys levels are needed to maximise profitability than for selling whole broilers. They also stated that prices of feed and breast meat affect the economic optimal Met + Cys levels only slightly. The conclusions of Pack and Schutte (1995) about the economics of Met + Cys levels are similar to the conclusions of the present study about the economics of balanced protein level.

Lys_{econ} versus Lys_{perf}

The economic model enables calculation of the balanced protein level at which gross margin is maximal (Lys_{econ}). However, it is common practise

among nutritionists to formulate diets that contain higher levels of balanced protein in order to maximise broiler performance (Lys_{perf}). Lys_{perf} for a grower diet (21-42 d) in the reference situation was 10.6 g/kg, calculated as Lys requirement for feed conversion (Chapter 4). Lys_{econ} for the grower diet in the reference situation was 8.9 g/kg for whole broilers. In this situation, feeding for maximum broiler performance ($Lys_{perf} = 10.6$ g/kg) instead of maximum profit ($Lys_{econ} = 8.9$ g/kg) decreases gross margin over the grower period by 2.65 €-cents per broiler. At higher prices of protein-rich raw materials (+20%), feeding for maximum broiler performance instead of maximum profit ($Lys_{econ} = 8.0$ g/kg; Table 3) would decrease profitability over the grower period even more (7.59 €-cents per broiler). For a typical broiler enterprise slaughtering 1 million broilers per week, each €-cent difference per broiler increases gross margin by 10.000 € per week. Thus, formulating diets for maximum profit instead of maximum broiler performance can result in considerable increases in profitability, particularly if the difference between Lys_{perf} and Lys_{econ} is more than 1.0 g/kg.

Implications

The results of this study imply that formulating diets either for maximum profit or for maximum performance results in different diets. Moreover, feeding for maximum profit instead of maximum broiler performance can strongly increase profitability of a broiler production enterprise. Once diets are formulated for maximum profit, only changes in age period, price of protein-rich raw materials and large changes in meat prices necessitate adaptation of the dietary balanced protein levels to maintain maximum profitability. Finally, the model simulations showed that for maximum profitability, dietary balanced protein level should depend on how the broilers are marketed; as whole birds, carcass or cut-up.

ACKNOWLEDGMENTS

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General Discussion

Physiologists are quite good at inventing reasons why some number is what it is observed to be, but considerably less good at predicting what it will be in the next case.

G.C. Emmans

The results of the studies described in the present thesis were discussed extensively in the discussion sections of each Chapter, and related to the objective in each Chapter. The contribution of each study to the overall objective of the dissertation was not yet reviewed. Therefore, the aim of this General Discussion is to evaluate to what extent the *overall objective* of this thesis is accomplished. Firstly, the contribution of the results of the first experiment, as described in Chapters 1 and 2, to existing broiler growth models is evaluated. It is then argued why existing growth models are less appropriate to realise the dissertation objective than was thought at the beginning of this study. Subsequently, it is discussed how the new developed growth model is better fit to fulfil the aim of the dissertation. An example is also given on how this model can be applied in practice. Finally, an overview is presented of the conclusions, implications and suggestions for further research.

The overall objective of this thesis was to develop a growth model for broiler chickens that could be easily used by practical nutritionists. The model should facilitate the selection of feeding strategies (in terms of dietary protein and energy) that results in the desired body composition of broilers while minimizing cost. A description of the genotype of a broiler chicken should be an input variable for the model.

CONTRIBUTION TO EXISTING BROILER GROWTH MODELS

The studies described in Chapters 1 and 2 were designed to validate two crucial theories that are commonly applied in growth simulation models. The first tested theory states that, at limiting protein intakes, protein deposition rate is independent of energy intake (Chapter 1). According to the other theory, fat-free body composition is independent of nutrition (Chapter 2).

Protein deposition rate

Several growth simulation models for pigs are based on the assumption that dietary protein (or amino acids) is preferentially used for protein deposition, unless energy availability or other factors (genotype or environment) become limiting (Whittemore and Fawcett, 1976; Moughan and Verstegen, 1988). It is postulated that, where protein intake is limiting, additional energy intake will not enhance protein deposition rate. The same hypothesis is used in growth simulation models for broilers (for

example in the EFG-model; Emmans, 1981), although it is hardly validated for this species.

The experiment described in Chapter 1 demonstrates indeed that, at limiting protein intake, additional energy intake has generally no effect on protein deposition rate in broilers. In this way, the experiment gives a more scientific basis to an important theory included in a broiler growth simulation model of about two decades ago (Emmans, 1981). In addition, the experiment in Chapter 1 supplies data on efficiency of protein utilization for protein deposition in two body weight ranges. Moreover, the experiment provides quantitative information on the use of energy for protein and lipid deposition in the carcass and organs fractions. These data on protein and energy utilization can be used to improve the accuracy of the protein and energy parameters in a broiler growth model.

Fat-free body composition

It is assumed in several growth models, that the relationships between water and protein, and between ash and protein, in the body of growing broiler chickens are independent of nutrition (Emmans, 1981; Black *et al.*, 1986). This theory was not supported by the study described in Chapter 2. It was concluded that simulation of chemical body composition in broilers solely based on protein deposition might be accurate in case of *ad libitum* access to balanced diets, but can induce systematic errors in the simulations in case of low feed intake levels or sub-optimal protein to energy ratios in the diet. Thus, for growth simulation under limiting nutritional conditions an alternative theory on water and ash growth in broilers should be developed. It should be noted, however, that any alternative theory would probably miss the attractive simplicity of the basic theory that fat-free body composition is independent of nutrition.

The study described in Chapter 2 delivered also estimates of the allometric relationships between water and protein, and between ash and protein, in the body of broiler chickens. It was shown, for example, that ash is at constant proportion of 15% of protein. The model of Emmans (1981) calculates ash as 16% of protein. The allometric relationship between water and protein in our study also differed from the one used by Emmans (1981). Thus, estimates of the allometric relationships by the present study can be used to update parameters in broiler growth models that include such allometric relationships.

During the present project, and for reasons discussed below, it was decided that mechanistic growth models in general are less appropriate to accomplish the dissertation objective. Therefore, the modifications that

should be made in the EFG-model (Emmans, 1981) or other mechanistic growth models for broiler chickens (for example, King, 2001), given the results of the experiments in Chapters 1 and 2, are not specified further.

Protein-free energy

An additional remark should be made on the definition of dietary energy in the experiments described in Chapters 1 and 2. In these experiment, dietary energy was defined as protein-free energy, which is energy coming from carbohydrates and fats only (Gerrits *et al*, 1996). This would make the two experimental variables (daily intakes of dietary energy and protein) independent of each other. Strictly speaking, this measure was effective since protein-free energy and protein are independent nutritional variables. However, it was not effective in solving the more important nutritional problem of dietary protein being unavoidably confounded with dietary energy available for maintenance and growth. As discussed in Chapter 1, part of the dietary protein is inevitably oxidised, thereby increasing the amount of energy available for maintenance and growth. Another disadvantage of defining energy as protein-free energy is that it is an uncommon energy scale in nutritional science in general and in poultry nutrition in particular. Consequently, the interpretation of the experiment (in terms of validation of nutritional theory) and the application to the practice of poultry nutrition is more complicated than if dietary energy would have been defined in a more common way, for example as AMEn. It can be concluded, therefore, that the advantages of defining dietary energy as protein-free energy instead of AMEn are less important than the disadvantages. The study described in Chapters 1 and 2 could have been done well with energy defined as AME instead of protein-free energy, like was done in studies of similar design with pigs (De Greef, 1992; Bikker, 1994).

LIMITATIONS OF EXISTING MECHANISTIC MODELS

The aim of our investigations was to develop a growth model for broiler chickens that could be easily used by practical nutritionists. In the General Introduction, it was argued that this model should be a mechanistic model. Mechanistic models have the advantage that these are more generally valid than empirical models (Black, 1995). Mechanistic models have a better change to be valid for extrapolation than empirical models, provided that the mechanisms of growth are represented in a correct way. In addition, mechanistic models can also include a theory on how broiler genotype affects nutritional responses (Emmans, 1981), which was a condition in the objective of this dissertation. However, during this

project we became more convinced that, if the final objective of the model is to serve as a tool for broiler nutritionists, existing mechanistic models do have two major limitations.

The first limitation concerns the type of body composition that is predicted. Mechanistic growth models until now generally predict the chemical body composition, defined in terms of protein, lipid, ash and water content of the body. Validated theories are available to predict the total chemical body composition from nutritional inputs (Chapters 1 and 2). For the practice of broiler meat production, however, it is not the chemical body composition but particularly the physical body composition (yields of carcass and breast meat, for example) that is of importance. In several modelling studies the partitioning of nutrients over physical body constituents was investigated (Bikker, 1994; Weis *et al.*, 2004; Chapters 1 and 2). However, there are no mechanistic theories available yet that predicts physical body composition from chemical body composition or directly from nutritional inputs (De Greef, 1995). Moreover, development of such a theory would be more complex than for chemical body composition. For chemical body composition, deposition of protein and energy (in protein and lipid) in the body can be predicted from protein and energy in the diet. Thus, input and output for such a theory can be stated on the same scales. This is not the case for a theory that predicts physical body composition from nutrition.

The second limitation concerns the prediction of feed intake. Mechanistic models often predict animal performance from daily nutrient intakes (Moughan *et al.*, 1987; Zoons *et al.*, 1991) and not directly from nutrient concentrations in the diet. This means that to apply such models for *ad libitum* fed animals (like broilers usually are in practice) an additional model would be needed to predict feed intake. The problem is that feed intake is difficult to predict accurately (Emmans, 1995), whereas the accuracy of model predictions is influenced largely by the accuracy of feed intake estimates (Black and De Lange, 1995). The EFG-model is a mechanistic model that predicts feed intake (Emmans, 1981). However, although the theory on feed intake in this model has some experimental support (Emmans, 1987), it has not been extensively tested for broiler chickens.

Thus, given the objective to develop a relevant tool for broiler nutritionists (physical body composition and appropriate for *ad libitum* feeding), existing mechanistic models are less fit to be used. On the other hand, models that are fully empirical are not appropriate as well, given the limited validity of this type of models (Black, 1995). Therefore, an intermediate type of model was developed in this study (Chapter 4). It is discussed below how this new

model, to some extent, combines the advantages of mechanistic and empirical models whereas it avoids some of the disadvantages of both types of models.

THE NEW MODEL IN RELATION TO THE DISSERTATION AIM

As mentioned before, there is no mechanistic theory available to predict the physical body composition of broilers from feed intake and diet composition. Therefore, the new model directly links physical body composition (carcass and breast meat yield) to dietary protein level. In this respect, the new model is empirical and not mechanistic. At the same time, and for two reasons, the new model is more generally valid than fully empirical models, like for example a dose-response curve obtained in a feeding trial (Chapter 3). Firstly, the absolute level of broiler performance is input instead of output of the new model. The relative responses that are predicted by the model appeared to be independent of the absolute performance level. Thus, the model predictions seem valid for a range of broiler performance levels. Secondly, the model is made more generally valid than fully empirical models by including additional variables: age, sex and year (Chapter 4).

Apart from the difficulties regarding physical body composition, most mechanistic models have the problem of accurately predicting *ad libitum* feed intake. Although the new model is developed specifically to simulate *ad libitum* feeding conditions, it does not directly predict feed intake. The new model predicts responses of *ad libitum* fed broilers to dietary protein level in terms of growth rate and feed conversion, but not for feed intake. Obviously, the response in feed intake can be calculated backwards from the predicted responses in growth rate and feed conversion. The advantage of this approach is that predicting dose-responses in growth rate and feed conversion to dietary protein is less complicated than predicting the same responses in feed intake. The mentioned responses in growth rate and feed conversion have a relatively simple exponential shape, whereas the shape of response in feed intake is much more complicated to describe mathematically (Figure 1).

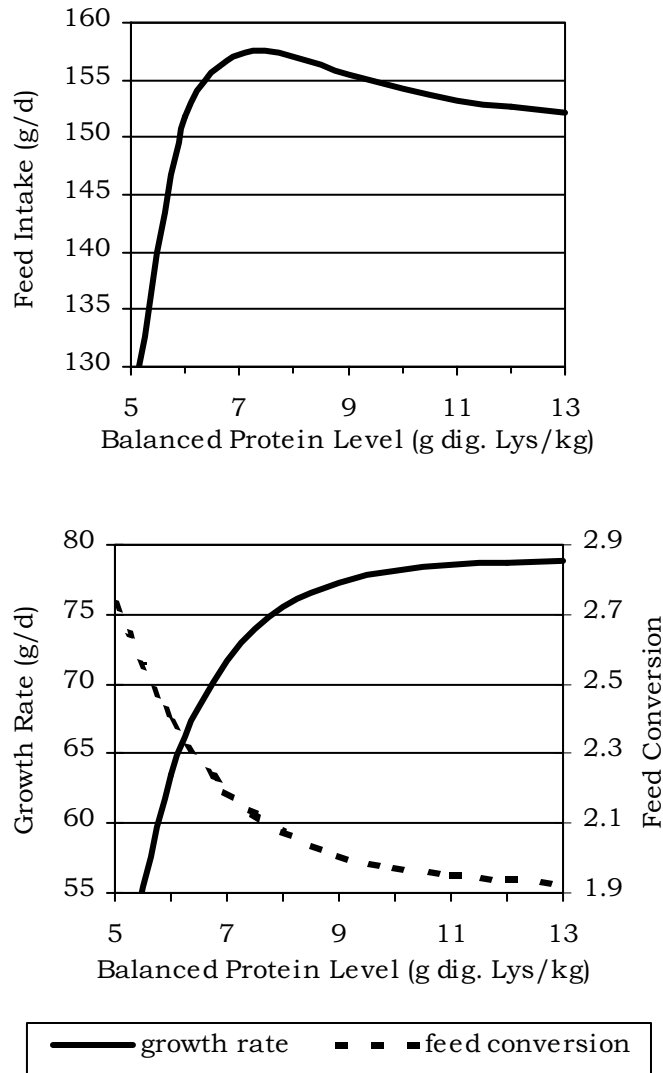


Figure 1. Shape of broiler responses to dietary balanced protein level, derived from the model on digestible Lys (Chapter 4). Responses in growth rate and feed conversion have a relatively simple exponential shape, whereas the shape of the response in feed intake is much more complicated to describe mathematically.

Some additional conditions were stated in objective of this dissertation. The intended model should also consider economic aspects. Moreover, a description of broiler genotype should be included as an input variable as well. The economic aspects were modelled in Chapter 5. The effect of broiler genotype was considered by the variable year, which represents the effect of genetic potential on dietary balanced protein requirement for maximising growth rate or minimizing feed conversion (Chapter 4). However, year was not included in the models for digestible lysine, nor in the models for body composition, because the data sets used for these models were all quite recent and comprised a too small range of 'years'. The effect of genetic potential on absolute level of broiler performance is not relevant for the model. The model predicts *relative* responses; that is, responses relative to the absolute (asymptotic) performance level. In our model, absolute level of predicted performance is input and not output.

In contrast to the dissertation objective, the model does not consider dietary energy. In Chapter 4, some reasons for not considering dietary energy were discussed.

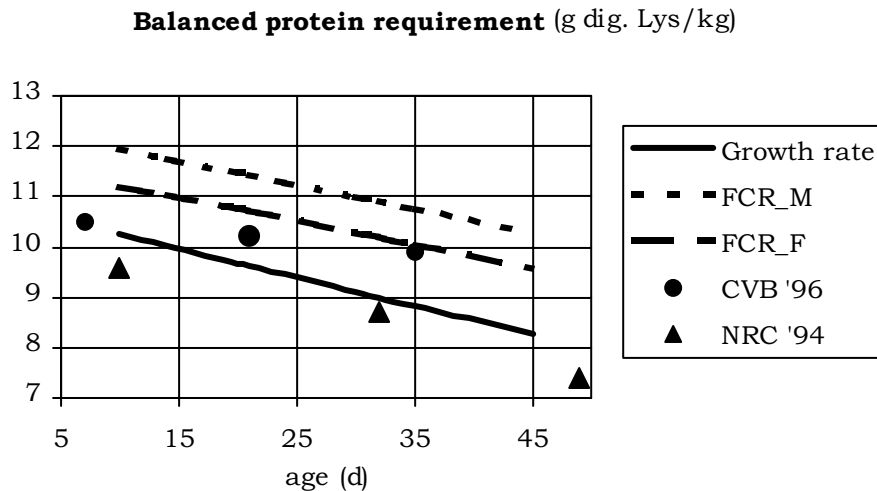


Figure 2. The effect of broiler age on balanced protein requirement. Model predictions on balanced protein requirement for maximum growth rate or for minimal feed conversion for males (FCR_M) and females (FCR_F). Model predictions derived from the model on digestible Lys in Chapter 4. Digestible Lys requirement estimates by NRC (1994) and CVB (1996) were included as a reference. NRC requirement estimates for digestible Lys were calculated as 87% of the requirement estimates for total Lys by NRC (1994).

EXAMPLE OF APPLYING THE MODEL IN PRACTICE

The model described in Chapter 4 predicts broiler responses to Dietary Balanced Protein level (DBP). According to this model, broiler responses to DBP can be seen as consisting of three parts: (i) the asymptotic performance level, (ii) the DBP requirement for maximum performance and (iii) the shape of the dose-response curve (that is, the rate of decrease in broiler performance if DBP is decreased below the DBP requirement). Asymptotic performance level is input for the model. It is taken from knowledge of maximum performance in practice. DBP requirement and the shape of the dose-response curve are predicted by the model. The model predictions on DBP requirement (g dig. Lys/kg) for growth rate and feed conversion are presented in Figure 2. As a reference, recommendations on Lys requirement by NRC (1994) and CVB (1996) are also included in Figure 2. The shape of the dose response curves for growth rate and feed conversion ratio, according to the model, are presented in Figure 3.

The dose-response relationships predicted by the model were used to analyse the economic aspects of DBP in broilers (Chapter 5). In addition, the model can be easily used by nutritionists to predict what would happen to broiler performance if modifications were made to the DBP level. This can be explained most easily by means of an example:

Example

A nutritionist considers decreasing the DBP level in the broiler diets, for example due to high prices of soy bean meal. Currently, the nutritionist is feeding male broilers a grower diet (21-42 days of age) with a DBP level of 10 g dig. Lys/ kg. He knows that the feed conversion over the grower period, using the present diet, is on average 1.90. The nutritionist plans to decrease DBP level to 8.5 g dig. Lys/kg. He wants to know what feed conversion he can expect with this new diet. The model predictions, as summarized in Figures 2 and 3, can easily solve the problem of the nutritionist.

Broiler age: The average broiler age is $((21+42)/2) = 31.5$ days of age.

DBP requirement: DBP requirement for minimising feed conversion of male broilers of 31.5 days of age is about 10.9 g Lys/kg (Figure 2).

Present diet: The present diet contains 10.0 g Lys/kg, which is 0.9 g Lys/kg (10.9 -/ - 10.0) below the requirement (RQ). The relative feed conversion at RQ-/- 0.9 is about 103.3% (Figure 3).

New diet: The new diet contains 8.5 g Lys/kg, which is 2.4 g Lys/kg (10.9 -/- 8.5) below the requirement (RQ). The relative feed conversion at RQ-/-2.4 is about 107.5% (Figure 3). The expected feed conversion at the new diet can now be calculated as $1.90 * (107.5/103.3) = 1.98$.

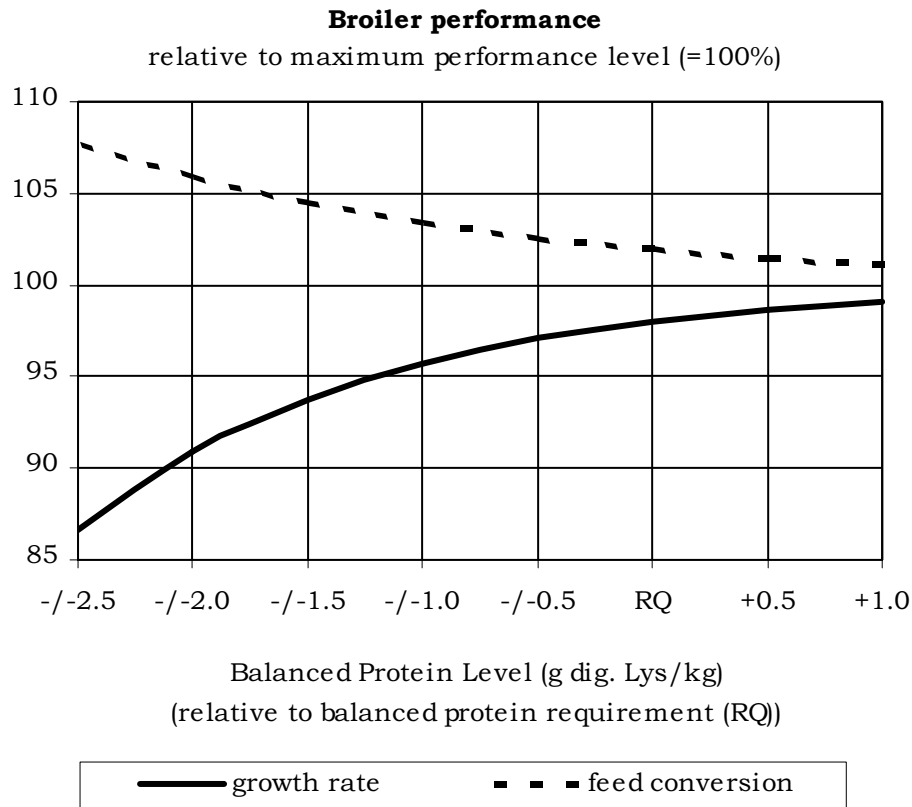


Figure 3. Broiler responses in growth rate and feed conversion (relative the asymptotic performance level (=100%)) to dietary balanced protein level (relative to the balanced protein requirement (RQ) for maximum growth rate or minimal feed conversion). These graphs are derived from the model on digestible Lys in Chapter 4 (model parameter C).

The problem in this example concerns a basic nutrition-related question, which cannot be answered by, for example, NRC (1994) and CVB (1996) recommendations. NRC and CVB recommendations (Figure 2) supply no information about differences in Lys requirement between sexes, or about differences in Lys requirement for optimising growth rate, feed conversion, carcass yield and breast meat yield, respectively. In addition, CVB (1996) recommendations, which are already higher than NRC (1994) recommendations (Figure 2), seems too low for modern broilers (Wijtten *et*

al., 2001; Lemme *et al.*, 2003). Another limitation is that NRC (1994) supplies no information on what happens to broiler performance if DBP is decreased below the requirement (= shape of the dose response curve). CVB (1996) presents information on the shape of dose response curves, but only for single amino acids and not for DBP levels.

In conclusion, the model gives an update on DBP requirements for modern broilers. This knowledge on DBP requirements is more detailed than NRC (1994) and CVB (1996) recommendations. In addition, the model supplies quantitative information on broiler performance at DBP levels *below* the DBP requirement. To our knowledge, it is the first time that this latter information is based on more than one experiment.

CONCLUSIONS AND IMPLICATIONS

The following main conclusions and implications follow from the research described in this dissertation:

- At limiting protein intake, additional energy intake has generally no effect on protein deposition rate in the body of broilers (Chapter 1). The related theory, which was initially developed for pigs, can thus be applied in broiler growth models as well.
- Fat-free body composition at a certain fat-free body weight is affected by feed intake level and by the protein to energy ratio in the diet (Chapter 2). This means that the independence of fat-free body composition from nutrition is not a valid assumption for broiler growth models.
- In broilers, ash weight is at a constant proportion of 15% of protein weight. With increasing protein weights, the amount of water per unit of protein decreases continuously; the allometric coefficient being 0.945 (Chapter 2). These facts are relevant to calibrate growth model parameters for modern broiler breeds.
- Broiler responses to dietary balanced protein (DBP) level depend on previous protein nutrition (Chapter 3). This suggests that DBP levels in grower and finisher diets should not be optimised independently, but simultaneously.
- The shape of dose-response curves describing broiler performance at varying DBP levels seems independent of the absolute level of broiler performance and of broiler age, sex and genetic potential

(Chapter 4). This constant shape of response, in combination with data on the DBP requirement (predicted by the model) and on the absolute level of performance (estimated by the model user himself) makes it possible to construct tailor-made dose-response curves without actual experimentation.

- DBP requirement for minimising feed conversion is 10 to 20% higher than for maximising growth rate or breast meat yield, and more than 30% higher than for maximising carcass yield (Chapter 4). DBP requirement for minimising feed conversion is higher than the Lys levels recommended by NRC (1994) and CVB (1996) (General Discussion). Thus, if the aim is to maximise broiler performance, higher amino acid levels are needed than suggested by NRC (1994) and CVB (1996).
- The shape of the dose-response curves in growth rate and feed conversion to DBP level is mathematically less complicated than the shape of the dose-response in feed intake to DBP level (General Discussion). This implies that it is easier to calculate the responses in feed intake from the responses in growth rate and feed conversion, than to model the responses in feed intake to DBP level directly.
- Formulating diets for either maximum profit or maximum broiler performance results in different DBP levels (Chapter 5). Formulating for maximum profit instead of maximum broiler performance may increase the profitability of a broiler production enterprise.
- The economic optimal DBP level for slaughtered broilers (not cut up) is lower than for cut up broilers, but higher than for live broilers (Chapter 5). This implies that for maximising profitability, DBP level should depend on how the broilers are marketed.

RECOMMENDATIONS FOR FURTHER RESEARCH

It can be concluded that the research described in this thesis realised the general objective to a large extent. Nevertheless, further research would be useful to validate the new model by means of independent dose-response studies; that is, dose-response studies not used for the development of the model. In Figure 4, simulations by the new model are compared with independent data from an experiment with male Hybro-G broilers (unpublished data from Nutreco Poultry & Rabbit Research Centre).

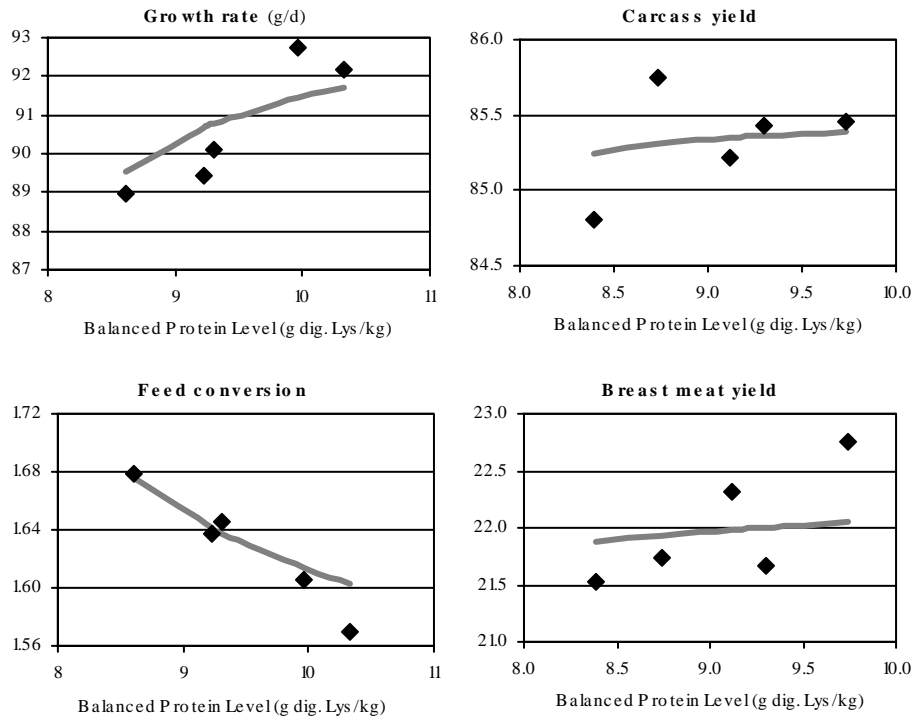


Figure 4. Broiler responses to dietary balanced protein level. Simulations by the new model compared with independent data (♦) from an experiment with male Hybro-G broilers. Growth rate and feed conversion responses during 21 to 35 days of age. Carcass yield (g/g body weight) and breast meat yield (g/g carcass) responses during 21 to 49 days of age.

The experimental data show variation among treatments that cannot be explained by the DBP level, particularly for the carcass and breast meat yield responses (Figure 4). This illustrates that it is difficult to obtain a reliable estimate of the shape of dose-response curves by means of just one or very few data sets. Consequently, it is impossible to validate the new model, which was based on 27 data sets from nine independent studies, by one experiment. Validation of the new model requires a set of dose-response experiments, including a wide range of DBP levels, with broilers varying in age, sex and genetic potential. Such experiments would also make it possible to include year as a variable in the models for digestible lysine as well as in the models for body composition.

Further research to investigate important assumptions in the model would be valuable as well. For example, on the hypothesis that difference in broiler weight at the end of an age period is maintained until slaughter age. The assumption that nutritional responses during an age period are

independent of previous nutrition is possibly an oversimplification (Chapter 3). Another aspect that deserves further attention is the possible difference in DBP requirement among commercial broiler breeds. One of our experiments showed, for example, that DBP requirement might be positively correlated with genetic potential for meat yield (Eits, 2001, Table 1 in Chapter 4).

Where models are intended to be used as a practical tool for nutritionists, one should be aware that further development of a model would not automatically make the model better fit for its aim. Any extensions to the model would make the model probably more complex, thereby running the serious risk that the model will not be applied in practice at all (Hilhorst, 1992). Moreover, any possible improvements in the model would require additional research time and money without giving the final answer, since the model would remain, by definition, a simplification of the nutritional reality. For those reasons, it is believed that tackling potential problems during implementation of the model predictions is of higher practical importance than further developing the model itself.

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Summary

INTRODUCTION

Protein is an important nutrient for growing broiler chickens, as it affects broiler performance, feed cost as well as nitrogen excretion. The effects of dietary protein in broiler diets depend on many interacting factors, like dietary energy level, broiler genotype and temperature. Consequently, choosing the optimal protein level for a broiler diet is a complex decision. A growth simulation model that evaluates these interacting factors simultaneously would be a useful tool in that respect. Several broiler growth models are available, which can be classified as empirical and mechanistic models. Empirical models work as a black box whereas mechanistic models describe underlying biological processes. An earlier study in our laboratory revealed that existing broiler growth models were inappropriate for being used as a practical tool for formulating broiler diets. Existing models were either too simple (empirical models) or too complex and inaccurate (mechanistic models).

Therefore, the objective of this dissertation was to develop a growth model for broiler chickens that could be easily used by practical nutritionists. The model should facilitate the selection of feeding strategies (in terms of dietary protein and energy) that results in the desired body composition of broilers while minimizing cost. A description of the genotype of a broiler chicken should be an input variable for the model.

Two important theories that are generally used in growth models were validated for broiler chickens (Chapters 1 and 2). Another general problem in growth models, namely the possible effects of early life nutrition on responses at later age, was investigated for protein nutrition in broilers (Chapter 3). A new model was developed that describes broiler responses to dietary balanced protein level (Chapter 4). Based on the predictions by this new model and data on feed and meat prices, the economic aspects of dietary balanced protein level in broiler diets were evaluated (Chapter 5).

PROTEIN DEPOSITION RATES IN BROILERS (CHAPTER 1)

It is an important assumption in several growth simulation models for pigs that dietary protein is preferentially used for protein deposition, unless energy availability or other factors (genotype or environment) become limiting. The key assumption here is that, where protein is limiting, protein

deposition rate will not increase with additional energy intake. There is hardly any experimental evidence that is relevant to the testing of the idea in broiler chickens.

Two experiments of similar design were conducted with male broiler chickens over two body weight ranges: 200 to 800 g (Experiment 1) and 800 to 1,600 g (Experiment 2). Each experiment consisted of 18 treatments: two levels of protein-free energy ($\text{energy}_{\text{pf}}$) intake, combined with nine protein to $\text{energy}_{\text{pf}}$ ratios (PE-ratio). Dietary protein was balanced for amino acid content. At the end of both experiments, broilers were divided into three fractions: carcass, organs and feathers. The carcass and organ fractions were analysed for protein, lipid, water and ash content.

Protein deposition rate increased with additional amino acid intake. No evidence was found that $\text{energy}_{\text{pf}}$ intake limited protein deposition rate at high amino acid intake. Where amino acid intake was limiting, additional intake of $\text{energy}_{\text{pf}}$ had generally no effect on protein deposition rate. It was concluded that the tested theory is valid to be used in broiler growth models.

FAT-FREE BODY COMPOSITION OF BROILERS (CHAPTER 2)

The independence of fat-free body composition from nutrition is another central assumption in most models that simulate animal growth. This assumption has not been extensively investigated. In this study, firstly, the allometric relationships of water and ash with protein were investigated in growing broiler chickens. Secondly, it was tested if the amounts of water or ash at a given protein weight were affected by nutritional factors. Analyses of the present study were based on the data obtained in the previous study on protein deposition rate.

The allometric relationships of water and ash with protein were different for carcass and organs. In broilers, ash weight was at a constant proportion of 15% of protein weight. With increasing protein weights, the amount of water per unit of protein decreased continuously; the allometric coefficient being 0.945 for carcass and organs together. The relationship between water and protein was not affected by nutrition, except for a 7% reduction in water weight at a fixed protein weight in the carcass in Experiment 1 at the lowest PE-ratio compared to the highest PE-ratio. The relationship between ash and protein was strongly affected by nutrition. The lowest PE-ratio increased ash weight at a fixed protein weight in the

carcass by up to 28%, compared to the highest PE-ratio. It was concluded that independence of fat-free body composition from nutrition is not a valid assumption for broiler growth models.

EFFECTS OF EARLY-LIFE PROTEIN NUTRITION ON LATER RESPONSES (CHAPTER 3)

If a model predicts responses of broiler chickens of a given age, the question arises to what extent the broilers responses depend on what has happened to the broiler at earlier age. A study was conducted with broiler chicks to test the effects of early life protein nutrition and sex on responses in growth and body composition to dietary balanced protein at later age. Effects on the incidence of metabolic disorders were also evaluated. From 11 to 26 d of age (EXP1), birds were fed eight diets varying in balanced protein to energy ratio (BPE-ratio) between 0.575 and 1.100 g digestible lysine per MJ AMEn. Birds from two treatment groups in EXP1 (BPE-ratio of 0.725 and 1.025 g/MJ, respectively) were subsequently used in a test from 26 to 41 d of age (EXP2). In EXP2, eight diets were fed varying in BPE-ratio between 0.500 and 1.025 g/MJ.

Responses in weight gain and feed conversion to BPE-ratio in EXP2 changed considerably when BPE-ratio in EXP1 was modified, irrespective of sex. Up to 10% improvement in both weight gain and feed conversion in EXP2 was observed if BPE-ratio in EXP1 was 0.725 compared to 1.025 g/MJ. With males, however, the effect of treatment in EXP1 on weight gain in EXP2 was present only at high BPE-ratios. For the relative gain of breast meat and abdominal fat, but not for carcass, the responses of male broilers to BPE-ratio in EXP2 were altered by the BPE-ratio in EXP1. With females, responses in composition of the gain to diet in EXP2 were independent of BPE-ratio in EXP1. The incidence of metabolic disorders was low, irrespective of treatment in EXP1. The lower BPE-ratio in EXP1 increased mortality in EXP2 from 0.8 to 3.6%. It was concluded that broiler responses to dietary balanced protein depend on previous protein nutrition and sex. Effects of early life protein nutrition on incidence of metabolic disorders were not observed. The results suggest that balanced protein levels in grower and finisher diets should not be optimised independently, but simultaneously.

A NEW MODEL ON BROILER RESPONSES TO DIETARY BALANCED PROTEIN (CHAPTER 4)

The ultimate aim of the present PhD-project was to develop a broiler growth model that should be fit to serve as a practical tool for nutritionists. During this project, we became aware that improving existing mechanistic broiler models would not be a feasible approach for this aim, because this type of models seems to be unfit to deal with physical body composition (for example, carcass, breast meat, abdominal fat) and to simulate *ad libitum* feed intake with acceptable accuracy and precision. Therefore, a new model was developed.

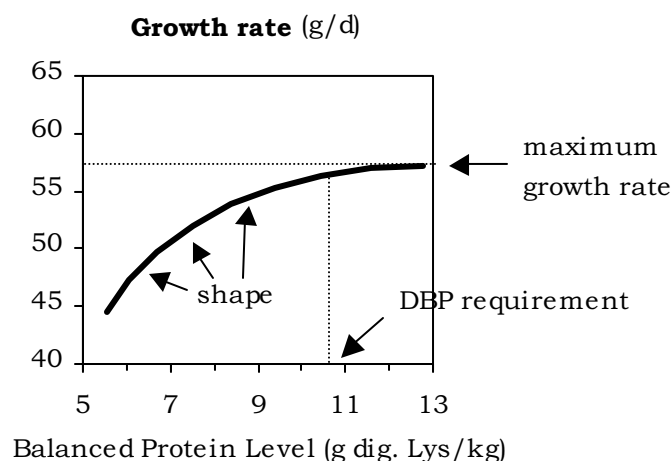


Figure 1. Hypothetical dose-response curve, describing broiler responses in growth rate to dietary balanced protein level (DBP). Dose-response curves can be seen as consisting of three parts: (1) maximum (asymptotic) performance level, (2) DBP requirement for maximum performance and (3) shape of the dose-response curve.

This model predicts broiler responses (growth rate, feed conversion ratio, carcass yield and breast meat yield) to dietary balanced protein (DBP) level. The model makes it possible to construct tailor-made dose-response curves without actual experimentation. In this new approach, dose-response curves are defined as consisting of three parts (Figure 1): the maximum (asymptotic) performance level, the DBP requirement (minimal DBP level to realise almost the asymptotic performance level) and the shape of the dose-response curves (rate of decrease in performance if DBP level is decreased below the DBP requirement). Asymptotic performance is input for the

model. The model predicts DBP requirement from broiler age, sex and year. Year indicates genetic potential. The shape of the dose-responses was shown to be constant, independent of asymptotic performance level, age, sex and year.

Broiler response studies from literature and the Nutreco Poultry Research Centre (27 data sets in total) were used in the model development to select significant variables, to quantify the parameters and to evaluate the accuracy of the predictions. Input variables were DBP level, asymptotic performance level, age, year (indicating genetic potential) and sex. The new model gave an accurate simulation of growth rate, feed conversion and breast meat for nearly all data sets. Accuracy was less for carcass. The new model is useful, among others, to evaluate the economic aspects of DBP in broilers.

THE ECONOMICS OF DIETARY BALANCED PROTEIN IN BROILERS (CHAPTER 5)

An economic model was developed that calculates economic optimal dietary balanced protein (DBP) levels for broiler chickens, based on technical input and prices of meat and feed (Figure 2).

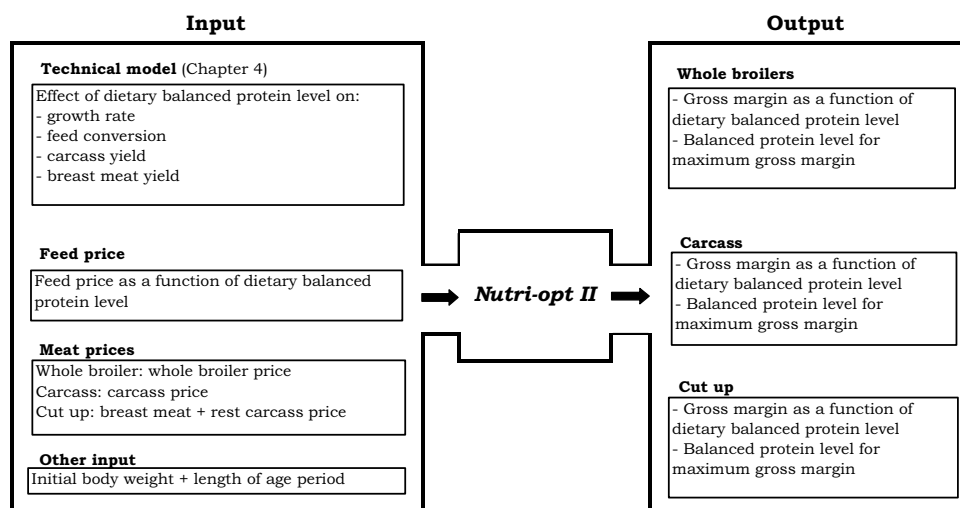


Figure 2. Overview of input and output for the economic model.

Technical input on broiler responses to DBP level (growth rate, feed conversion, carcass yield and breast meat yield) was obtained from the new model described above. Changes in broiler age, price of protein-rich raw materials and large changes (40%) in meat prices result in economic relevant differences in DBP level for maximum profit. Effects of changes in sex or feed price on DBP level for maximum profit are negligible. Model simulations also showed that formulating diets for maximum profit instead of maximum broiler performance results in different diets. Feeding for maximum profit instead of maximum broiler performance may strongly increase profitability of a broiler production enterprise. Model simulations revealed as well that DBP level for maximum profitability depend on how the broilers are marketed; as whole birds, carcass or cut up.

CONCLUSIONS AND IMPLICATIONS

The following main conclusions and implications follow from the research described in this dissertation:

- At limiting protein intake, additional energy intake has generally no effect on protein deposition rate in the body of broilers (Chapter 1). The related theory, which was initially developed for pigs, can thus be applied in broiler growth models as well.
- Fat-free body composition at a certain fat-free body weight is affected by feed intake level and by the protein to energy ratio in the diet (Chapter 2). This means that the independence of fat-free body composition from nutrition is not a valid assumption for broiler growth models.
- In broilers, ash weight is at a constant proportion of 15% of protein weight. With increasing protein weights, the amount of water per unit of protein decreases continuously; the allometric coefficient being 0.945 (Chapter 2). These facts are relevant to calibrate growth model parameters for modern broiler breeds.
- Broiler responses to dietary balanced protein (DBP) level depend on previous protein nutrition (Chapter 3). This suggests that DBP levels in grower and finisher diets should not be optimised independently, but simultaneously.

- The shape of dose-response curves describing broiler performance at varying DBP levels seems independent of the absolute level of broiler performance and of broiler age, sex and genetic potential (Chapter 4). This constant shape of response, in combination with data on the DBP requirement (predicted by the model) and on the absolute level of performance (estimated by the model user himself) makes it possible to construct tailor-made dose-response curves without actual experimentation.
- DBP requirement for minimising feed conversion is 10 to 20% higher than for maximising growth rate or breast meat yield, and more than 30% higher than for maximising carcass yield (Chapter 4). DBP requirement for minimising feed conversion is higher than the Lys levels recommended by NRC (1994) and CVB (1996) (General Discussion). Thus, if the aim is to maximise broiler performance, higher amino acid levels are needed than suggested by NRC (1994) and CVB (1996).
- The shape of the dose-response curves in growth rate and feed conversion to DBP level is mathematically less complicated than the shape of the dose-response in feed intake to DBP level (General Discussion). This implies that it is easier to calculate the responses in feed intake from the responses in growth rate and feed conversion, than to model the responses in feed intake to DBP level directly.
- Formulating diets for either maximum profit or maximum broiler performance results in different DBP levels (Chapter 5). Formulating for maximum profit instead of maximum broiler performance may increase the profitability of a broiler production enterprise.
- The economic optimal DBP level for broilers to be sold as carcass (not cut up) is lower than for broilers to be sold as cut up, but higher than for broilers to be sold as live broilers (Chapter 5). This implies that for maximising profitability, DBP level should depend on how the broilers are marketed.

Samenvatting

INLEIDING

Eiwit is een belangrijke voedingstof voor vleeskuikens. Vleeskuikens hebben vooral eiwit nodig om te kunnen groeien. Daarnaast is eiwit nodig om gezond te blijven, bijvoorbeeld voor een goede afweer tegen ziekten. De kwaliteit van een voer voor vleeskuikens hangt daarom sterk af van de hoeveelheid eiwit in het voer en van de kwaliteit van dit eiwit. De kwaliteit van het eiwit is goed als het goed verteerbaar is door het kuiken. Daarnaast dienen de verschillende eiwitbestanddelen (aminozuren) in de juiste verhouding aanwezig te zijn. Eiwit met de juiste verhouding aan aminozuren wordt vaak 'ideaal eiwit' of 'gebalanceerd eiwit' (balanced protein) genoemd.

Over de gewenste eiwitkwaliteit is al veel kennis beschikbaar. Ingewikkelder is de keuze van de juiste *hoeveelheid* (gebalanceerd) eiwit. De hoeveelheid voedereiwit heeft namelijk veel verschillende effecten. Zo beïnvloedt het bijvoorbeeld de groeisnelheid van vleeskuikens en ook de voederconversie. Voederconversie is de hoeveelheid voer die nodig is om een kuiken 1 kg te laten groeien. Ook de hoeveelheid vet in het kuiken en de beveesdheid, bijvoorbeeld het aandeel borstvlees (kipfilet), hangen af van de hoeveelheid eiwit in het voer. Daarbij komt dat al deze effecten van voedereiwit moeilijk zijn te voorspellen, omdat ze afhangen van het vleeskuikenras, de staltemperatuur, de hoeveelheid energie in het voer, en vele andere factoren. Dit probleem is niet op te lossen door veiligheidshalve voeders te maken met een extra hoog eiwitgehalte. Voer met veel eiwit erin is duur. Bovendien, de overmaat aan eiwit in het voer wordt uitgescheiden en belast zo het milieu.

In de praktijk zijn het de nutritionisten die de grondstofsamenstelling van vleeskuikenvoeders bepalen, en daarmee ook het eiwitgehalte van het voer. Om een keuze van het eiwitgehalte te kunnen maken, dient het hierboven geschetste, ingewikkelde probleem te worden versimpeld. Anders gezegd, er moet een model gemaakt worden. In de literatuur zijn verschillende modellen voor vleeskuikenvoeding beschreven. Probleem met deze bestaande modellen is dat ze te ingewikkeld, of juist te simpel zijn, om te dienen als hulpmiddel voor nutritionisten. Ingewikkelde modellen hebben vaak als probleem dat voor de berekeningen gegevens nodig zijn die een

nutritionist in de praktijk niet beschikbaar heeft. Het model dient ook weer niet te simpel te zijn. Het model moet het mogelijk maken om betere beslissingen te nemen dan op basis van bestaande kennis.

Het doel van dit proefschrift was om een eenvoudig model te maken waarmee nutritionisten een betere beslissing kunnen nemen over de hoeveelheid eiwit in vleeskuikenvoeders. Het model moest rekening houden met de erfelijke eigenschappen van het kuiken. Ook diende het model de economische consequenties te berekenen, bijvoorbeeld hoe de voerkosten en de winst per vleeskuiken afhangen van het eiwitgehalte in het voer.

Als eerste is voor twee belangrijke theorieën over diervoeding die in veel modellen worden toegepast, onderzocht in hoeverre deze geldig zijn voor vleeskuikens (Hoofdstukken 1 en 2). Vervolgens is onderzocht hoe de eiwithoeveelheid in het voer voor jonge kuikens van invloed is op de reactie van kuikens op het eiwitgehalte in het voer op latere leeftijd (Hoofdstuk 3). In Hoofdstuk 4 is het technische model beschreven, dat voorspelt hoe vleeskuikens presteren op voeders met verschillende eiwitgehalten. Tenslotte is het economische model ontwikkeld waarmee de economische consequenties van het eiwitgehalte in vleeskuikenvoer berekend kunnen worden (Hoofdstuk 5).

DAGELIJKSE EIWITAANZET IN VLEESKUIKENS BIJ VERSCHILLENDE VOEDERS (HOOFDSTUK 1)

Chemisch gezien bestaat een kuiken uit vier bestanddelen: eiwit, vet, water en as. Zo bekeken, betekent groei dus dat het dier extra eiwit, vet, water en as aanzet in het lichaam. Wanneer we de dagelijkse aanzet van deze vier bestanddelen kunnen voorspellen, hebben we daarmee een manier (model) om de groeisnelheid van het dier te voorspellen. Verschillende groeimodellen voor dieren zijn gebaseerd op deze theorie over chemische lichaamssamenstelling.

Een belangrijke aanname in dergelijke groeimodellen is dat voedereiwit met name wordt gebruikt voor de groei van eiwit in het lichaam, en veel minder wordt gebruikt als energiebron. Als de dagelijks opgenomen hoeveelheid voedereiwit onvoldoende is, zal het dier minder eiwit in het lichaam aanzetten. Deze modellen gaan er vanuit dat in dit laatste geval de

dagelijkse eiwitaanzet niet kan worden verhoogd door extra voederenergie (koolhydraten en vetten) te verstrekken. Deze theorie blijkt te kloppen voor varkens, maar niet voor kalveren. Voor vleeskuikens was onvoldoende bekend of deze theorie klopt.

Om de genoemde theorie te toetsen voor vleeskuikens, zijn twee proeven uitgevoerd met vleeskuikenhanen, voor twee gewichtstrajecten: van 200 tot 800 gram (proef 1) en van 800 tot 1600 gram (proef 2). Elke proef bestond uit 18 verschillende behandelingen: negen verschillende voeders (die verschilden in de verhouding tussen het eiwit- en energiegehalte) in twee verschillende dagelijkse hoeveelheden. Aan het einde van elke proef werden de kuikens geslacht en opgedeeld in drie delen: karkas, organen en veren. Van karkas en organen werd de chemische samenstelling bepaald: eiwit, vet, water en as.

De dagelijkse aanzet van eiwit in het lichaam bleek inderdaad toe te nemen als de kuikens meer voedereiwit kregen. In geval de opname aan voedereiwit de dagelijkse aanzet van eiwit beperkte, kon de eiwitaanzet niet worden verhoogd door extra voederenergie te verstrekken. Kortom, de kuikens deden wat we verwachtten. Op basis van deze resultaten is daarom de conclusie getrokken dat de geteste theorie inderdaad gebruikt kan worden in groeimodellen voor vleeskuikens.

WATER EN AS IN RELATIE TOT EIWIT IN HET LICHAAM VAN VLEESKUIKENS (HOOFDSTUK 2)

Zoals hierboven al genoemd, heeft voer grote invloed op de aanzet van eiwit in het lichaam. Ook de aanzet van vet hangt sterk af van het voer. Groeimodellen gaan er vaak vanuit dat de aanzet van de overige twee lichaamsbestanddelen, water en as, niet direct afhangt van het voer. Deze theorie stelt dat de hoeveelheid water en as alleen afhangt van de hoeveelheid eiwit in het lichaam, onafhankelijk welk voer de dieren krijgen. Ook deze theorie, die voor vleeskuikens nauwelijks is onderzocht, is getoetst met de gegevens van de proef uit Hoofdstuk 1.

Als eerst is berekend hoeveel water en as aanwezig was in het lichaam van vleeskuikens, en dit is vergeleken met de hoeveelheid eiwit in het lichaam. Het bleek dat de hoeveelheid as altijd 15% was van de hoeveelheid eiwit.

Voor water lag dit wat ingewikkelder. Hoe groter de hoeveelheid eiwit in het lichaam, hoe minder water er, relatief ten opzichte van eiwit, in het lichaam was. Anders gezegd, hoe zwaarder de kuikens, hoe 'droger' de groei.

Als tweede is nagegaan of deze relatie tussen as en eiwit, en tussen water en eiwit, inderdaad niet afhing van het voer dat de kuikens kregen. De relatie tussen water en eiwit was inderdaad meestal niet afhankelijk van het voer. Alleen in proef 1 was er een effect van voer op deze relatie. De relatie tussen as en eiwit was, in beide proeven, sterk afhankelijk van het voer. Bij een bepaalde hoeveelheid eiwit kon de hoeveelheid as wel 28% verschillen, afhankelijk van welk voer de kuikens kregen. De conclusie is daarom getrokken dat de hoeveelheid water en as in het lichaam van vleeskuikens niet alleen afhangen van de hoeveelheid eiwit in het lichaam, maar ook afhangen van het voer.

VOEDEREIWIT OP JONGE LEEFTIJD: GEVOLGEN OP LATERE LEEFTIJD (HOOFDSTUK 3)

Met een model is het mogelijk te voorspellen wat het effect van voeding is op de prestaties van vleeskuikens van een bepaalde leeftijd. De vraag is dan wel in welke mate deze prestaties afhangen van de voeding op jongere leeftijd. Om deze vraag te beantwoorden voor wat betreft het eiwitgehalte in het voer is een proef uitgevoerd met vleeskuikens. In deze proef zijn zowel hanen als hennen gebruikt. Kuikens van 26 tot 41 dagen leeftijd kregen één van de in totaal acht verschillende voeders. De voeders verschilden in de verhouding tussen het eiwit- en het energiegehalte. De kuikens hadden tijdens de voorafgaande leeftijdsfase (11 tot 26 dagen) een eiwitrijk voer, of juist een eiwitarm voer gehad. Gemeten is in hoeverre de reacties van de kuikens op het voer in de tweede leeftijdsfase afhingen van het voer in de voorafgaande leeftijdsfase. Ook is onderzocht of de voeding op jonge leeftijd invloed had op de gezondheid (afwezigheid van metabole stoornissen) op latere leeftijd.

Als het eiwitgehalte van het voer op jonge leeftijd laag was, presteerden de kuikens op latere leeftijd beter. Dit gold zowel voor hanen als voor hennen. In geval van een eiwitarm voer op jonge leeftijd, waren de groeisnelheid en de voederconversie op latere leeftijd tot 10% beter dan wanneer op jonge

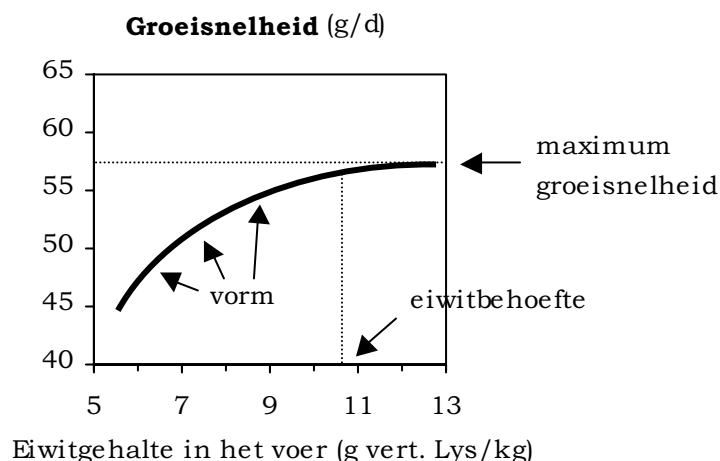
leeftijd een eiwitrijk voer was verstrekt. De hanen vertoonden deze compensatie in groei alleen indien het eiwitgehalte in het voer op latere leeftijd relatief hoog was. De samenstelling van de groei (bijvoorbeeld het aandeel karkas, borstvlies en buikvet) van hennen op latere leeftijd was niet afhankelijk van het voer op jonge leeftijd. Bij hanen had het voer op jonge leeftijd wel effect op de samenstelling van de groei op latere leeftijd, maar alleen voor wat betreft borstvlies en buikvet. Metabole stoornissen waren in alle proefgroepen nagenoeg afwezig. Er konden daarom geen conclusies worden getrokken over het effect van voer op het voorkomen (en voorkomen) van metabole stoornissen. De kuikensterfte op latere leeftijd nam af van 3,6% tot 0,8%, indien op jonge leeftijd een eiwitrijk voer in plaats van een eiwitarm voer werd verstrekt. De resultaten van deze proef suggereren dat het eiwitgehalte van de voeders voor respectievelijk jongere en oudere kuikens, niet los van elkaar gezien kunnen worden.

EEN NIEUW MODEL VOOR EIWIT IN VLEESKUIKENVOER (HOOFDSTUK 4)

Het uiteindelijke doel van dit promotieonderzoek was om een model te maken dat geschikt was om te dienen als praktisch hulpmiddel voor nutritionisten. Tijdens dit onderzoek bleek dat aanpassing van bestaande modellen geen goede manier was om deze doelstelling te realiseren, en wel om twee redenen. Ten eerste voorspellen de bestaande modellen vaak de chemische lichaamssamenstelling van kuikens (eiwit, vet, water en as) terwijl in de praktijk juist de anatomische lichaamssamenstelling (bijvoorbeeld, het aandeel karkas, borstvlies en buikvet) van belang is. Ten tweede is bij bestaande modellen een betrouwbare voorspelling van de voeropname essentieel; iets wat nog niet goed mogelijk is. Om deze redenen is een nieuw model ontwikkeld.

Het nieuwe model voorspelt het effect van het eiwitgehalte in het voer op de prestaties van vleeskuikens (groeisnelheid, voederconversie en het aandeel karkas en borstvlies in het lichaam). De relatie tussen bijvoorbeeld groeisnelheid en het eiwitgehalte kan in een grafiek worden weergegeven (Figuur 1). Een dergelijke relatie wordt wel dosis-respons curve genoemd. Eerst is een wiskundige formule opgesteld die de dosis-respons curve opdeelt in drie fasen (Figuur 1): (1) de maximale groeisnelheid (de asymptoot), (2) de eiwitbehoefte (dat is het eiwitgehalte dat minimaal nodig

is om de maximale groeisnelheid te realiseren) en (3) de vorm van de curve (de mate waarmee de groeisnelheid afneemt wanneer het eiwitgehalte wordt verlaagd beneden de eiwitbehoefte).



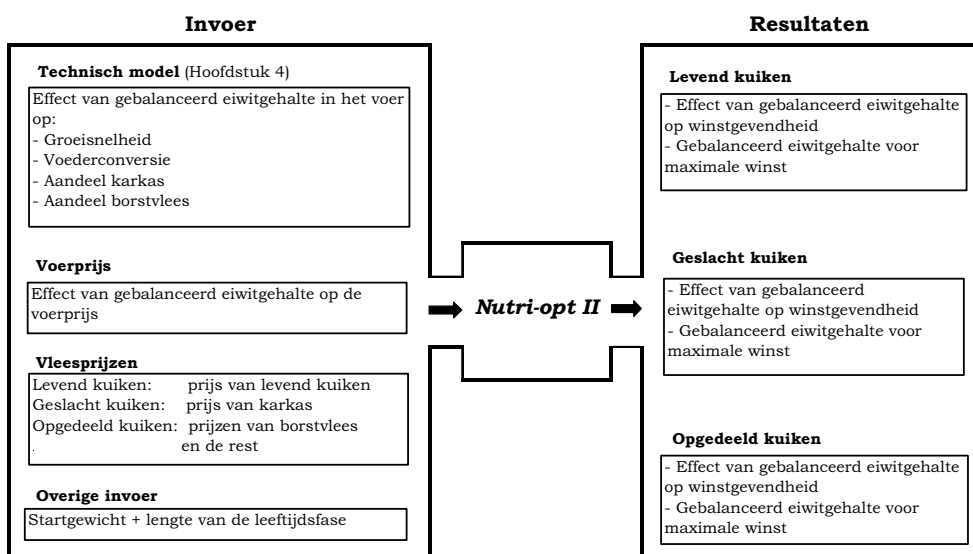
Figuur 1. Voorbeeld van een dosis-respons curve die de relatie beschrijft tussen groeisnelheid van vleeskuikens en het (gebalanceerde) eiwitgehalte in het voer. De dosis-respons curve kan worden opgedeeld in drie delen: (1) de maximale groeisnelheid, (2) de eiwitbehoefte voor maximale groeisnelheid en (3) de vorm van de dosis-respons curve.

Bovenstaande betekent dat de nutritionist met gegevens over de maximale groeisnelheid, de eiwitbehoefte en de vorm van de curve, de totale dosis-respons curve kan beschrijven. De dosis-respons curve maakt het vervolgens mogelijk om te voorspellen hoeveel de groeisnelheid zal wijzigen bij een verandering in het eiwitgehalte. Het eerstgenoemde, de maximale groeisnelheid, is doorgaans goed bekend bij nutritionisten. Zo niet, dan kan deze worden ingeschat op basis van gegevens van de fokkerijorganisatie. Het tweede, de eiwitbehoefte, hangt af van onder andere de leeftijd en sekse van de kuikens, alsook van de erfelijke eigenschappen. Daarom is een formule ontwikkeld die de eiwitbehoefte voorspelt op basis van leeftijd, sekse en erfelijke eigenschappen. Het derde, de vorm van de curve, bleek onafhankelijk van het absolute niveau van de kuikenprestaties. Anders gezegd, de relatieve afname in prestaties van de kuikens bij verlaging van het eiwitgehalte bleek hetzelfde voor bijvoorbeeld jonge en oudere kuikens.

Het model is ontwikkeld op basis van resultaten van proeven uit de literatuur en van proeven op het pluimveeonderzoekscentrum van Nutreco. Dit model, in combinatie met een economisch model, maakt het mogelijk om de economische gevolgen van verschillende eiwitgehalten in vleeskuikenvoeders door te rekenen.

ECONOMISCHE ASPECTEN VAN EIWIT IN VLEESKUIKENVOER (HOOFDSTUK 5)

Tenslotte is een economisch model ontwikkeld: 'Nutri-opt II'. Dit model berekent hoe de voerkosten, opbrengsten en winstgevendheid per vleeskuiken afhangen van het eiwitgehalte in het voer. Middels Nutri-opt II kan dus bijvoorbeeld worden uitgerekend bij welk eiwitgehalte de winstgevendheid maximaal is. Voor deze berekeningen heeft het model verschillende invoergegevens nodig (Figuur 2).



Figuur 2. Overzicht van het economische model "Nutri-opt II": invoergegevens en modelvoorspellingen.

Ten eerste zijn berekeningen nodig van het model uit Hoofdstuk 4 om te kwantificeren hoe de vleeskuikেনprestaties (groeisnelheid, voederconversie

en het aandeel karkas en borstvlees) afhangen van het eiwitgehalte in het voer. Daarnaast zijn gegevens nodig over vleesprijzen en voerprijzen. De gegevens over voerprijs betreft informatie over de relatie tussen voerprijs en het eiwitgehalte in het voer.

Simulaties met het economische model toonden aan dat veranderingen in leeftijd van de kuikens of in prijs van de eiwitrijke voedergrondstoffen, of grote veranderingen in vleesprijzen (40%) resulteren in economisch relevante verschillen in het eiwitgehalte voor maximale winstgevendheid. Effecten van sexe en algemeen prijsniveau van het voer waren in dat opzicht verwaarloosbaar. Modelsimulaties lieten verder zien dat het eiwitgehalte sterk kan verschillen, afhankelijk of de voeders worden geformuleerd voor maximale winst of juist voor maximale vleeskuikenprestaties. Formuleren van de voeders voor maximale winst in plaats van voor maximale vleeskuikenprestaties kan de winstgevendheid beduidend verbeteren. Tenslotte maakten de modelsimulaties duidelijk dat het eiwitgehalte voor maximale winst afhangt van hoe de vleeskuikens worden verkocht; als levend kuiken, geslacht kuiken of opgedeeld kuiken (borstvlees en overige delen).

CONCLUSIES EN PRAKTISCHE IMPLICATIES

De volgende conclusies en praktische implicaties volgen uit dit promotieonderzoek:

- Wanneer de eiwitopname beperkend is, heeft extra energieopname geen effect op de dagelijkse eiwitaanzet in het lichaam van vleeskuikens (Hoofdstuk 1). De hieraan gerelateerde theorie, die oorspronkelijk was ontwikkeld voor varkens, kan dus ook worden toegepast in groeimodellen voor vleeskuikens.
- De relatie tussen water en as enerzijds en eiwit anderzijds in het lichaam van vleeskuikens hangt af het voer (Hoofdstuk 2). Dit betekent dat de desbetreffende theorie over de vetvrije lichaamssamenstelling niet geldig is voor vleeskuikens.
- Daar waar de hoeveelheid eiwit in het lichaam van vleeskuikens toeneemt tijdens de groei, blijft de hoeveelheid as constant op 15%

van de hoeveelheid eiwit, terwijl de hoeveelheid water per eenheid eiwit continue afneemt (Hoofdstuk 2). Middels deze en andere gegevens uit Hoofdstuk 2 kan een groei-model voor vleeskuikens worden aangepast aan de moderne vleeskuikenrassen.

- Het effect van het eiwitgehalte in het voer op de prestaties van vleeskuikens hangt af van het eiwitgehalte in het voer gedurende de voorafgaande leeftijdsfase (Hoofdstuk 3). Dit suggereert dat de eiwitgehalten in voeders voor respectievelijk jonge en oudere kuikens, niet los van elkaar gezien kunnen worden.
- De vorm van de dosis-respons curve, die de relatie beschrijft tussen vleeskuikenprestaties en het eiwitgehalte in het voer, lijkt onafhankelijk van het absolute niveau van de vleeskuikenprestaties (Hoofdstuk 4). Deze constante vorm, in combinatie met gegevens over de eiwitbehoefte (voorspeld door het model) en het absolute prestatieniveau (ingeschat door de nutritionist), maakt het mogelijk voor een nutritionist om de te voorspellen hoe de vleeskuikenprestaties afhangen van het eiwitgehalte in het voer, zonder dat daarvoor proeven nodig zijn.
- De eiwitbehoefte om de beste voederconversie te realiseren is 10 tot 20% hoger dan voor de maximale groeisnelheid of het hoogste aandeel borstvlees, en meer dan 30% hoger dan nodig is voor het hoogste aandeel karkas (Hoofdstuk 4). De eiwitbehoefte voor de beste voederconversie is hoger dan de behoefte aan aminozuren zoals geadviseerd door het NRC (1994) en het CVB (1996) (General Discussion). Wanneer het doel is om maximale vleeskuikenprestaties te realiseren, lijken derhalve hogere aminozuurgehalten in het voer nodig dan volgens het NRC (1994) en het CVB (1996).
- Voor wat betreft de reacties van het kuiken op het eiwitgehalte van het voer geldt, dat de vorm van de dosis-respons curven voor groeisnelheid en voederconversie, wiskundig gezien, minder gecompliceerd is dan de vorm van de dosis-respons curve voor voeropname (General Discussion). Dit impliceert dat het gemakkelijker is om de curve voor voeropname te berekenen uit de

curven voor groeisnelheid en voederconversie, dan om de curve voor voeropname direct te modelleren.

- Het eiwitgehalte in vleeskuikenvoeders kan sterk verschillen, afhankelijk of de voeders worden geformuleerd voor maximale winst of voor maximale vleeskuikenprestaties (Hoofdstuk 5). Formuleren van de voeders voor maximale winst in plaats van voor maximale vleeskuikenprestaties kan de winstgevendheid beduidend verbeteren.
- Het economisch optimale eiwitgehalte in het voer voor kuikens te verkopen als karkas (niet opgedeeld) is lager dan voor kuikens te verkopen als opgedeeld kuiken (opgedeeld in borstvlees en overige vleessoorten), maar hoger dan voor kuikens die levend worden verkocht (Hoofdstuk 5). Dit impliceert dat het eiwitgehalte voor maximale winstgevendheid afhangt van hoe de vleeskuikens worden vermarkt.

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Curriculum Vitae

Ruud Eits (doopnaam: Rudolf Maurits Eits) is op 6 mei 1975 geboren te Zeist. Zijn VWO diploma behaalde hij in 1993 aan het Van Lodenstein College te Amersfoort. In 1993 begon hij de studie Zoötechniek – specialisatie veevoeding - aan de Universiteit van Wageningen. In 1997 liep hij drie maanden stage aan het Scottish Agricultural College in Edinburgh. Tijdens deze stage participeerde hij in een project voor modellering van de melkgift bij koeien. Ook was hij tijdens zijn studie in Wageningen twee jaar parttime werkzaam als biologiedocent aan een middelbare school. De studie in Wageningen sloot hij in 1997 *cum laude* af met als afstudeervakken veevoeding en onderwijskunde. Op dat moment was hij reeds werkzaam bij UTD mengvoeder te Maarssen als nutritionist en onderzoeker pluimvee. Sinds de overname van UTD mengvoeder door Nutreco in 1998 werkt hij als onderzoeker voor het Nutreco Poultry and Rabbit Research Centre nabij Madrid. Sinds 1997 heeft hij, naast andere R&D projecten, gewerkt aan het promotieonderzoek. Hij zal zijn werkzaamheden in de huidige functie voorzetten.

Ruud Eits (Christian name: Rudolf Maurits Eits) was born on the 6th of May in 1975 in Zeist, The Netherlands. In 1993 he graduated from secondary education. In the same year he started the study Animal Science at Wageningen University. In 1997 he went for 3 months to Scotland for his practical training at the Scottish Agricultural College in Edinburgh. For this training, he participated in a dairy cow-modelling project. During his study in Wageningen, he was part-time employed as Biology teacher at a secondary school. In 1997, he graduated *cum laude* for his masters after MSc research in Animal Nutrition and in Didactics. At that time, he was already working as poultry nutritionist and researcher for UTD mengvoeder, a Dutch animal nutrition company. Since the acquisition of UTD by Nutreco in 1998, he has been working as a researcher at the Nutreco Poultry and Rabbit Research Centre near Madrid, Spain. Since 1997 he has been working on the PhD-project and other R&D projects. He will continue his work in the present job.

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