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EVALUATING A BROILER GROWTH MODEL

C. FISHER¹ and R.M. GOUS²Summary

Simulation models of broiler growth are useful both as scientific and commercial tools. The needs of these two areas can only be reconciled if the model is a 'glass' box, with the contents of the model being transparent and available to the user. Models are only representations of reality and some evaluation has to be done each time the model is used.

Evaluation can usefully be carried out in the following steps: 1. Consider the bounds of the model; 2. Evaluate the theoretical structure of the model; 3. Test the model against existing experimental data; 4. Evaluate the predictions of the model in practical applications.

Various aspects of evaluating the EFG Broiler Growth Model (EFG Software, Natal, S.A.) are presented in this paper.

I. INTRODUCTION

Proving the commercial value of predictive models used in broiler production is obviously essential; the more so if the model is itself a commercial product. Model evaluation is a multi-step process involving not only those who develop (and sell) the model but also its users. Critically, the process of evaluation will depend on the type of model under consideration.

The model discussed here (the EFG Broiler Growth Model) implements a theory of growth and feed intake in broilers outlined by Emmans and Fisher (1986) and developed further by its main author G.C. Emmans (1987, 1995). The principal features of the model are as follows: *Firstly*; potential growth, a genetically determined characteristic, is defined by three growth and one feathering parameter. A Gompertz curve relating the potential for feather-free body protein growth to post-hatching time is the central feature. Potential body fatness is also seen as being under genetic control. *Secondly*; by assuming that birds have a purpose - to achieve their potential growth of body protein - a general theory of *ad libitum* feed intake can be elaborated (Emmans, 1997). *Thirdly*; by analysing how the environment, both physical and nutritional, will prevent a bird achieving its potential, actual performance under defined conditions can be computed. *Fourthly*; nutritional transactions (energy and amino acids only) are considered in conventional ways except that food energy is computed as 'effective' energy (Emmans, 1994) and not in the more limited way as ME. *Fifthly*; by using these principles the model simulates the growth of body and feather protein, fat, water and ash over successive intervals of time with daily summary of outputs. At each stage the composition of the body is computed using equations based on allometry with body protein weight. Biological and economic indexes of performance allow the results to be assessed. Evaluating a theory-based model of this sort raises a dichotomy. On the one hand there is a scientific need to prove the model 'wrong' so that it can be improved. Conversely there is a need to prove the model 'right' so that it may be used. This contradiction may be resolved by seeing the model as a 'glass' box, rather than the usual 'black' box, so that users of the model can share fully the understanding of what it does and what it does not do. If models are to develop in both a scientific and commercial sense then such openness seems to be essential.

¹ EFG Software (Natal), Leyden Old House, Kirknewton, Midlothian, EH27 8DQ, U.K.

² Dept of Animal and Poultry Science, University of Natal, Pietermaritzburg, South Africa.

II. MODEL EVALUATION

(a) Model bounds

Consideration of what the model includes and excludes is a useful first step in evaluation. Factors included are the genotype of the bird, including feathering; flock characteristics such as stocking density, mortality, distribution of mortality in time, and economic parameters; environmental variables temperature and humidity; diet composition in terms of energy and amino acids and the use of controlled feeding. Feed form, mash versus pellets, is partly implemented. Important factors that are not considered at this stage of development include: response to temperature stress, diurnal variations in temperature and lighting. With a theory based model desiderata for inclusion of a factor include not only its importance but also the availability of a proper theoretical treatment that is compatible with the rest of the model.

(b) Validating the theory

Evaluating and testing the theory that a model uses, independently of evaluating the overall model, is an essential process. Only in this way can the model be continuously improved. Models which become so complex that the component hypotheses cannot be independently tested may cease to be useful from a scientific point of view.

Potential feather-free body protein growth - a Gompertz curve: the assumed Gompertz curve, relating body protein, Bp, to time, is the central element of the growth theory of Emmans (1987, 1995). The curve defines two of the four genetic parameters in the model and through derived allometric relationships determines other body components. The assumption that a bird seeks to achieve its potential allows *ad libitum* feed intake to be simulated.

The essential assumption is that potential growth in Bp follows a smooth, continuous path which can be described empirically by a growth curve. The Gompertz curve is chosen because it represents the available data better than alternative curves, because of its mathematical properties leading to derived allometric relationships for all body components and because its parameters, although largely empirical, can be visualised and estimated experimentally. A frequent error in growth modelling is to apply such curves to observed growth data without applying the caveat that environmental conditions must be non-limiting. Under limiting conditions there is no inherent reason why growth should be continuous or smooth with respect to time, nor follow any single empirical curve. Hruby *et al.* (1996) describe protein growth of modern broiler strains under putative non-limiting conditions created by choice-feeding. They found that the Gompertz curve described their data very effectively and was statistically superior to the other forms examined (logistic and polynomial). The parameters of the Gompertz curve can be estimated from suitable data sets on immature birds (see Hancock *et al.*, 1995 for an example).

The definition of energy transactions in terms of 'effective energy' (Emmans, 1994) gives the model an important source of strength. In comparison with ME, effective energy (EE) takes account of diet composition (crude fibre, fat, digestibility) on energy utilisation. Few data sets are available to test the calculation of feed intake using the ideas of the effective energy system. Håkansson *et al.* (1978) present one set for broilers (see Emmans, 1994) which gives strong support to the theory. However use of these data to test the model is only partly justified since they were used by Emmans to quantify one of the parameters used in the prediction system. The turkey data in Figure 1 (Fisher and Emmans, 1992) provide a valid test of the system.

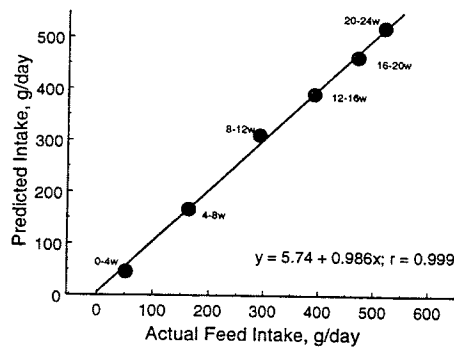


Figure 1. Actual feed intake and intake predicted using the effective energy scale in growing turkeys. The data points refer to successive periods of growth in male birds grown on a conventional high-energy feed (Fisher and Emmans, 1992)

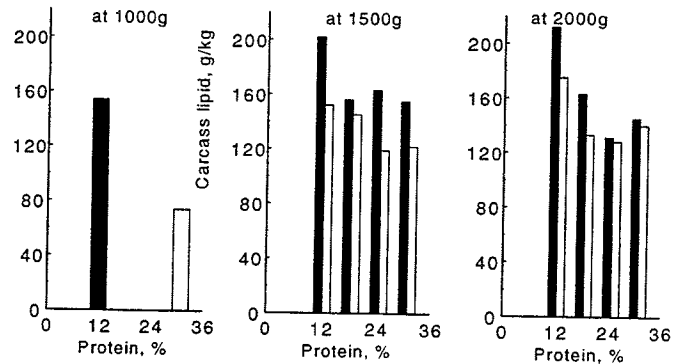


Figure 2. Changes in carcass lipid in broilers given different crude protein levels (Gous *et al.*, 1992).

The idea that birds strive to achieve their potential is best examined by looking at changes in fatness following a change in diet composition. Gous (1995) described an experiment to test this aspect of the model at this meeting. A single result is shown in Figure 2. Birds were made fat or lean at 1kg by feeding low and high protein levels. A range of protein levels was then given to each group during a recovery period. It can be seen that the lipid content of the fat birds did not change greatly after 1kg, implying a much lower lipid content in the growth. The lean birds however showed increasing fat content. The fatness of both groups reflected the protein content given during re-alimentation but by 2kg both groups had the same lipid content when given a sufficient level of protein. These data are consistent with the theory used in the model that birds will strive to achieve an inherent body composition determined by their genotype.

(c) Validating the model: comparison with experiments

Comparison of model predictions with the results of experiments provides the most readily available method of evaluating a model. Logically all possible experiments of a given type should be tested but this is time consuming work and such an ideal has not been achieved. The examples presented here were selected because they were good experiments and described in the literature in sufficient detail. In preparing this paper no experiments have been rejected because they failed to show the model in a good light.

This type of comparison raises many complex issues and is subject to many difficulties both theoretical and practical. The main difficulties are: *Firstly*; the available description of the experiment will not contain all the information required by the model. Estimates must be used. *Secondly*; the observed performance may, unknowingly, be influenced by factors which lie outside the bounds of the model. In these circumstances calibration of model output simply to make them look like the experimental results is not justifiable. *Thirdly*; the performance of the birds may not, in fact, wholly reflect the variables that the experimenter thought were under control. This is probably a common feature of nutrition experiments. A special case is that factors that are held constant, especially the environment, may interact with the treatments being used; such interactions are not revealed by the experiment but may influence model calculations.

Such points are discussed below in the context of individual experiments. Comparison of the results of a lysine response study with model predictions is shown in two ways in Figure 3. In Figure 3A predicted performance across treatments is correlated with the experimental results. This type of comparison is widely used in modelling, especially when the variation in the real-world results is not subject to experimental control. The interpretation of such a comparison rests on the magnitude of the correlation and the goodness-of-fit to the line $x = y$. Both these may be examined formally but the statistical testing of such hypotheses is the subject of considerable controversy (Thornton and Hansen, 1996; Colson *et al.*, 1995). This will not be discussed here.

A less formal test is to compare the response to the experimental variables as observed and as predicted by the model. (see Figures 3B,C,D). This is more useful for the present purpose and this type of graph is mostly used in this paper. At this stage formal statistical comparisons have not been made although these could be devised, for example by comparing residual standard deviations.

(d) Amino acid response studies

Han and Baker (1993) describe responses to lysine level in broilers from days 22 to 43. The experimental results and their reproduction using the model are shown in Figure 3. For gain-over-food (FCE) there is a very close correlation between the observed and model results in both males and females (Figure 3A). The responses in FCE are also quite well reproduced, especially in males (Figure 3B). These results test those components of the model concerned with nutrient utilisation; these are generally strong. A more severe test is to look at the components of FCE separately as this requires that responses over time and *ad libitum* food intake are simulated.

In males the simulation of the growth response is quite successful (Figure 3C). Simulated birds are slightly larger at low lysine levels and, more significantly, show declining body weight at lysine levels above 0.8%. The pattern of food intake response described by the model is considerably different from the experiment, with higher intakes at low lysine levels and a much steeper decline at higher levels. In females there is a similar pattern of results but overall the comparison is less good than in the males (Figure 3D).

This experiment thus shows a reasonable agreement between the model and the experiment, but more importantly, it reveals the serious limitations of this type of test and also, what can be learned about nutrition experiments by using a model. In simple terms, the theory of the model envisages that the bird attempts to eat enough food to meet its lysine requirement. Thus at low lysine levels food intake will increase and the birds will be fatter. The model tries to analyse what factors prevent the bird eating enough food to reach its potential and, in general and in this case, the ability to lose heat will normally be the limiting factor. Han and Baker (1993) do not describe the environmental temperature used in this experiment. But the birds were kept in cages in a temperature controlled building and for the model exercise described here it was assumed that the effective temperature experienced by the birds was constant at 21°C. This assumption is quite critical but fine-tuning is not really worthwhile since the effective temperature experienced by a bird on deficient feed may be different from that on an adequate feed. The deficient bird will certainly be 'colder' and may well modify its behaviour, and thus the effective temperature, as a result. Such issues will profoundly affect the results obtained in an experiment, but they are usually ignored. It is difficult to see how they can be taken into account in an exercise of the kind being discussed here.

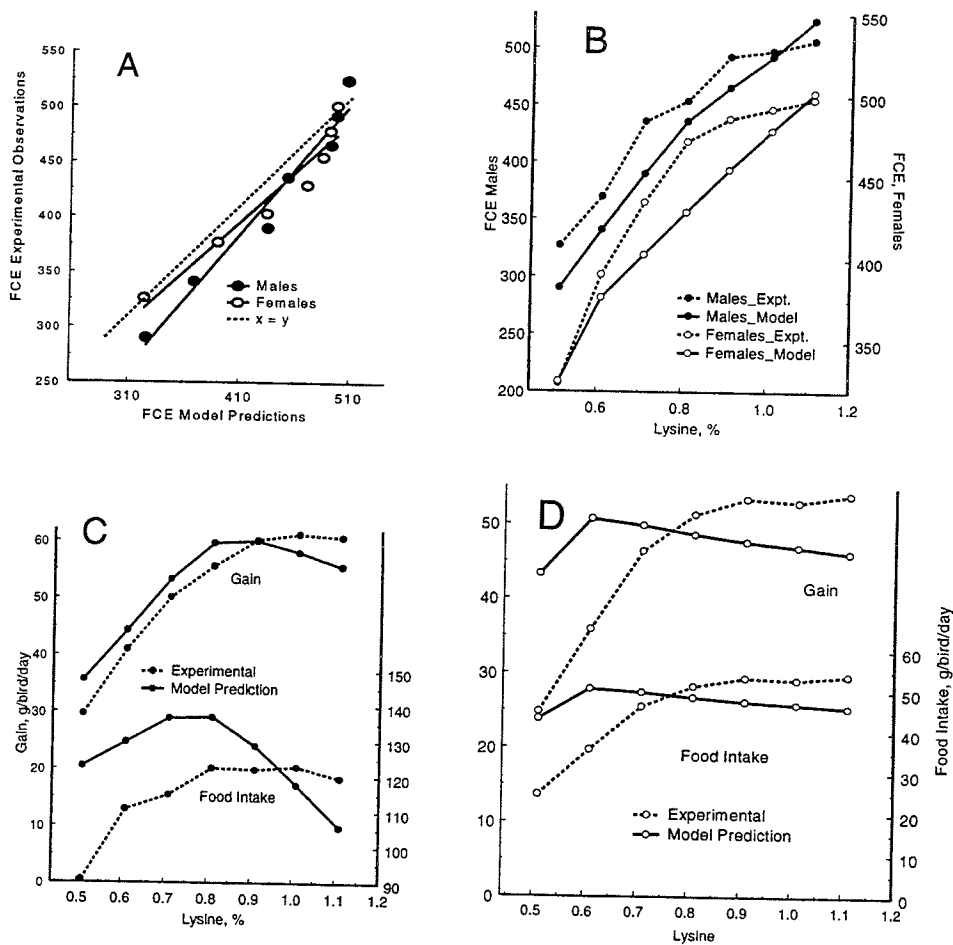


Figure 3 Responses to lysine in broilers aged 22-43d. Data from Han and Baker (1994). For simulation the model was calibrated to give similar growth rates in males at the lysine level which was just adequate. Birds were kept in cages and the room temperature was set at 21°C. Diets as described by the authors with some additional calculation. A. Correlation of observed and predicted FCE. B. Responses in FCE. C. Gain and food intake, males. D. Gain and food intake, females.

At the higher levels of lysine, where commercial decisions are likely to be made, this temperature effect will probably be quite small. However at these higher levels the model predicts reductions in food intake, and fatness, as lysine increases. As fatness reduces, the birds tend to become a bit smaller since maximum gain of other body components has already been reached (Figures 3C and 3D). These effects were not seen in the experimental data. Some approximate calculation of the response in terms of the fat-free body show that differences in fatness account entirely for the lack of agreement between the model and the experiment in this area of the response (these data are not shown here). The most likely explanation is that the experimental feeds became limiting in some other nutrient and the birds continued to eat for this in spite of having sufficient lysine. Such a nutrient may be another amino acid or a mineral or vitamin. The use of the model encourages the consideration of such possibilities and the improved design of experimental feeds.

(e) Responses to environmental temperature

The response of broilers to variations in environmental temperature presents the most severe challenge to modellers. Apart from cold-thermogenesis, which can probably be ignored in practice, heat is produced as a result of maintenance and activity, protein and lipid growth, digestion and nitrogen excretion (Emmans, 1994). Heat loss to the environment at a given effective temperature will reflect feathering, posture, blood flow, panting and so on. Effective temperature will be determined by factors such as radiant input, conductive losses, humidity, air movement and diurnal variations.

The integration of this complexity into a single model has made quite a lot of progress (e.g. Bruce and Clark, 1979) and is more widely applied in pig models (Moughan, *et al.*, 1995) than in current broiler models. However whilst the modelling is feasible it has not been demonstrated that the considerable increase in complexity will lead to improved commercial predictions. Specifically the management of broilers at high temperatures, in excess of about 31°C, may be too complex to benefit directly from modelling.

The EFG Broiler Growth Model has a simple set of rules about the effect of temperature. A single effective temperature (and humidity) is provided as input for each day. Heat loss calculations take account of humidity, feathering, stocking density and are scaled for a given genotype and degree of maturity. Heat production calculations are based on the effective energy scheme (Emmans, 1994) and take account of a wide variety of factors. Specifically the model does not contain concepts of either physiological or behavioural adaptation by the bird to temperature and thus it is not a high temperature model. Limitation of heat loss is the most common factor determining food intake so the model reflects very sensitively the various inputs. Even with a simple set of rules model behaviour is often very complex in this area. Simulation of a temperature experiment is illustrated in Figure 4. Charles *et al.* (1981) studied post-brooding temperatures in the range 15-27°C and also used four dietary nutrient density levels and broilers of both sexes. The scatter diagram in Figure 4a shows that the overall correlation between observed and simulated responses was reasonably close. The overall pattern of response in FCE (Figure 4b) is also well reproduced by the model, given the complexity of this experiment. Separate consideration of growth and feed intake (data not shown) reveals that the response of both components to temperature is well predicted. However the response to diet composition is less successfully simulated.

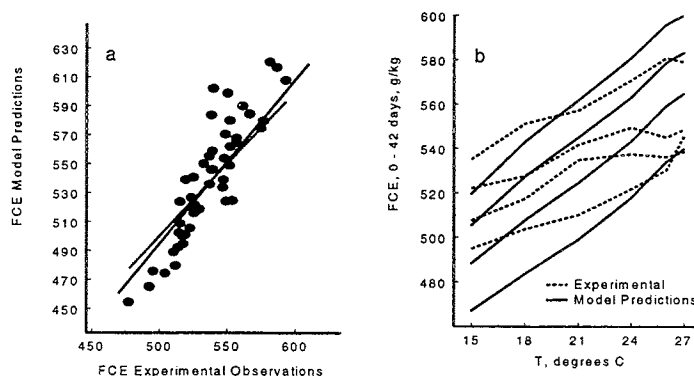


Figure 4. Effect of temperature and diet nutrient density on broiler performance. Data from Charles *et al.* (1981); mixed sex broilers reared in environmental chambers to 49 days. The model was not calibrated to the data set.

The model predicted that the four feeds used would support rather similar growth rates with wide variations in feed intake. The birds themselves responded by keeping intake fairly constant across feeds but showing wide differences in growth. The possible reasons for this difference between the experiment and the model are legion and speculation is not really rewarding. However it should be noted that the predicted model response is consistent with most nutritional experiments of this sort (Fisher and Wilson, 1974) whilst this experiment showed an unusual pattern of response.

(f) Responses to dietary energy and nutrient density

Leeson *et al.* (1996a) describe two experiments in which dietary energy (Exp. 1) and both energy and protein (Exp. 2) were diluted with a mixture of sand and oat hulls. Extensive dilution, up to 50%, was used, so this experiment provides a good test for a simulation model. Comparison of experimental and model results is shown in Figure 5. In general there is good agreement between the two. Food intake tended to be overestimated by the model; this may have reflected factors such as temperature, feeding space or an overestimate of the feeding value of oat hulls or an underestimate of the moisture content of the sand. None of these factors was defined in the paper describing the experiment. The model predicted much larger responses in body fatness than were indicated by the data reported for abdominal fat pad weight (data not shown).

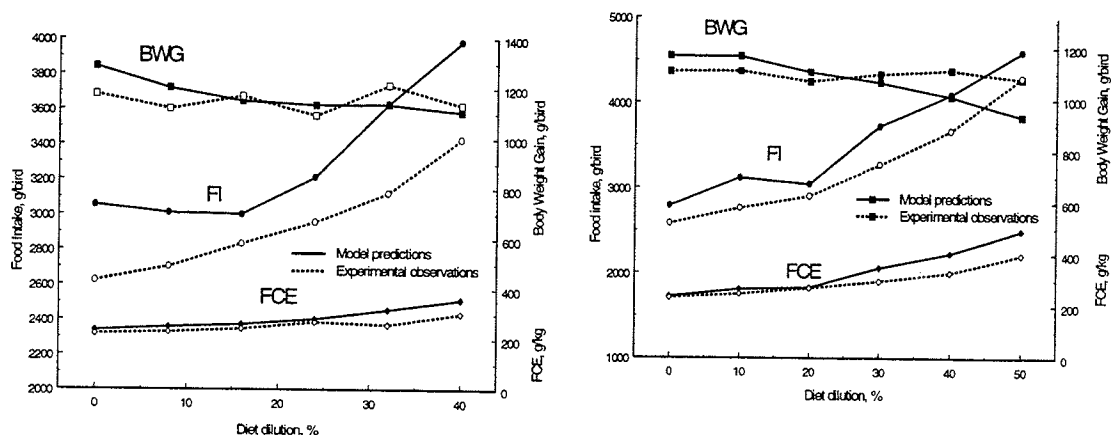


Figure 5. Effect of diet dilution on broiler performance (Leeson *et al.*, 1996a). Diet compositions were recalculated to provide the information required by the model. The model was not calibrated to the data set. Left-hand figure; Exp.1 in which energy was diluted. Right-hand figure; Exp 2 in which both energy and protein were diluted.

A more classical energy-protein experiment was described by Pesti and Fletcher (1983). This provides a useful test because it involves an old genotype and thus illustrates the capacity of the model to deal with genetic change. Also a very narrow range of response was reported so the test is a precise one. The experimental and model predictions for FCE and body fatness are shown in Figure 6. For FCE the agreement is very close both in pattern and amplitude of response. For fatness there is less close agreement although part of the pattern is reproduced. In general the model sees adjustment of body fatness as a larger component of

response than the birds revealed. There are many possible reasons for this: temperature is not well defined and some features of diet design may also have influenced the actual experimental results.

The correlation of experimental and model results for the FCE data shown in Figure 6 was 0.904, being highly significant. For growth and feed intake considered separately the correlations were negligible. This again emphasises that the prediction of the rates of growth processes over time is much more difficult than predicting nutrient utilisation.

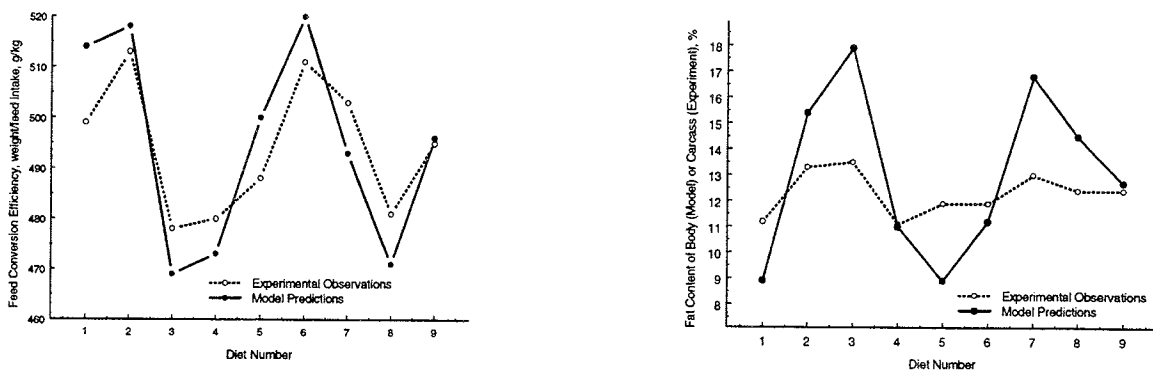


Figure 6. Response to energy and protein in broilers. Data from Pesti and Fletcher (1983). The model was adjusted to a 1970's genotype (see Oldham *et al.* 1997) and diet compositions were re-calculated.

(g) Controlled feeding

Controlled or restricted feeding is becoming a common feature in the broiler industry in Europe and user control of intake is a necessary feature of the model. Since the prediction of the rate of intake over time is the most challenging problem tackled in the model it is to be expected that removal of this element will increase considerably the accuracy with which the model reflects the real world.

Leeson *et al.* (1996b) report experiments in which four dietary energy levels were fed either *ad libitum* or according to a controlled intake pattern. The effect of feed restriction on growth and feed intake is well represented by the model and Figure 7b suggests that the low feed intake (and hence high conversion) observed at the highest energy level when fed *ad libitum* might be an artefact. In all cases the response to energy is also well simulated. The overall effects of the treatments on feed conversion (Figure 7c) are very small and the small differences between the model and the experiment probably reflect only such issues as temperatures and feathering. The reduction in fatness with declining energy content was overestimated somewhat in the *ad libitum* fed birds with almost exact agreement under controlled feeding (Figure 7d). This emphasises that the difficult prediction is of feed intake over time and all the factors which affect it, whilst the close agreement under controlled feeding suggests that the rules for nutrient intake and the resultant partitioning are about correct.

(h) Using the model

This section on model use is included under the heading 'Model Evaluation' to stress the importance of continuing evaluation by the model user and of the scientific transparency of the model.

Experience has shown many applications of the EFG Broiler Growth Model. These range from it being used as a starting point in the development of 'in-house' models by large companies to a wide range of direct applications in broiler nutrition and production decision making. The most common use is alongside R & D programmes where a question is simultaneously addressed by the model and by an experiment. Such application is analogous to the retrospective analyses presented above and offers the following advantages over reliance on experiments alone. *Firstly*; there is much greater understanding about the interactions between the three components involved – genotype, feed and environment. These are all inevitably involved in the outcome of an experiment. *Secondly*; treatment options can be evaluated before the experiment starts, giving a better choice of treatments. *Thirdly*; probable underlying biological changes e.g. in body composition, can be followed in detail. *Fourthly*; assessment of a wider range of biological outcomes can easily be made, and *fifthly*; economic assessment of the results under different scenarios can be easily done.

For some types of decision the use of experiments may be either impossible or too constrained. The use of a model then has additional value. Some examples from recent experience illustrate this. 1. A broiler production company normally slaughtered large male birds at 55 days. With some frequency market conditions dictated a delay in slaughtering of 7-10 days. How should the birds be fed during such a 'holding' period? Use of the model allowed a wide range of options to be tested. 2. A broiler company with no R & D facilities was able to re-direct policy on amino acid levels with great benefits. 3. Experiments on feeding programmes, i.e. the sequence and timing of feeds, are difficult simply because so many options are available and because interactions with other factors are likely to be important. An example of using the model is shown below. 4. Restricted or controlled feeding experiments are also very complex owing to the almost unbounded range of options concerning timing and degree of restriction. Unravelling this complexity with a model has had great commercial benefit.

Saleh *et al.* (1997) report a typical 'feeding programme' experiment. Starter feed was given for 7, 14 or 21 days and a finisher feed from 21, 28 35 or 42 days to broilers killed at 56 days. All $4 \times 3 = 12$ options were tested. The biological effects of the treatments were very small and reproduction of this experiment in the model is complex and not very easy. However the correlation result in Figure 8a suggests reasonable agreement between the experimental and model results. Figure 8b shows body lipid growth on the two treatments which gave the fattest (starter 0-14, finisher 21-56 days) and the leanest birds (starter 0-21; finisher 42-56 days). This additional information about the changes taking place during the experiment clearly enhances its value considerably. Economic calculation within the model allows the results to be assessed under different scenarios and for the assessment of the results to be made at different ages.

III. DISCUSSION

It seems to be important to develop modelling in the scientific community if progress is to be made. For this purpose models will have to be open. It is suggested here that developers of commercial models can both benefit, and make a more useful contribution to the poultry industry, if they take a similar approach.

It is not possible here to discuss the many areas where further development of both theory and modelling technique is required, nor to list the approaches which have been or might be useful. Undoubtedly a central problem in simulation modelling is the need to consider populations of birds rather than a single 'average' bird. The structure of the EFG Broiler Growth Model facilitates this in the way that genotypes are described (see Emmans and Fisher, 1986) and some examples are given by Gous (1997, in press). Similar issues in pig modelling have been discussed by Ferguson *et al.* (1997).

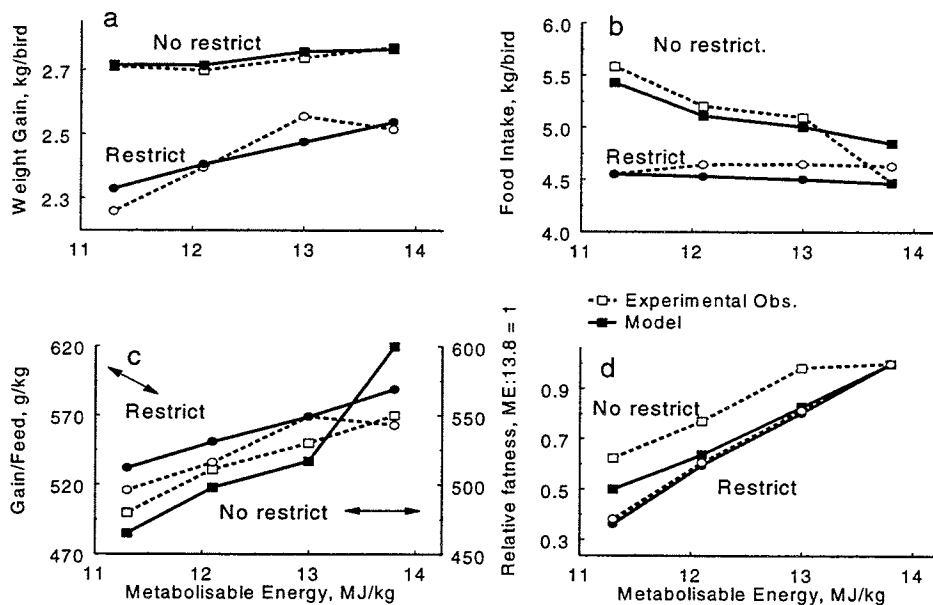


Figure 7. Restricted feeding and dietary energy level in broilers. Data from Leeson *et al.* (1996b). Model predictions made without calibration.

It is clear that different parts of any model will have different strengths. In the type of model discussed here the description of nutrient utilisation and growth, once intake is known, is relatively strong or 'hard'. The prediction of food intake over time is much more difficult and presents more challenging problems.

Systems of broiler production are being developed in which real-time data on bird growth and feed intake are continuously available in addition to control of inputs. The creation of models to use such data and to provide sophisticated methods of optimising production in such systems should be a much simpler task than the one discussed here.

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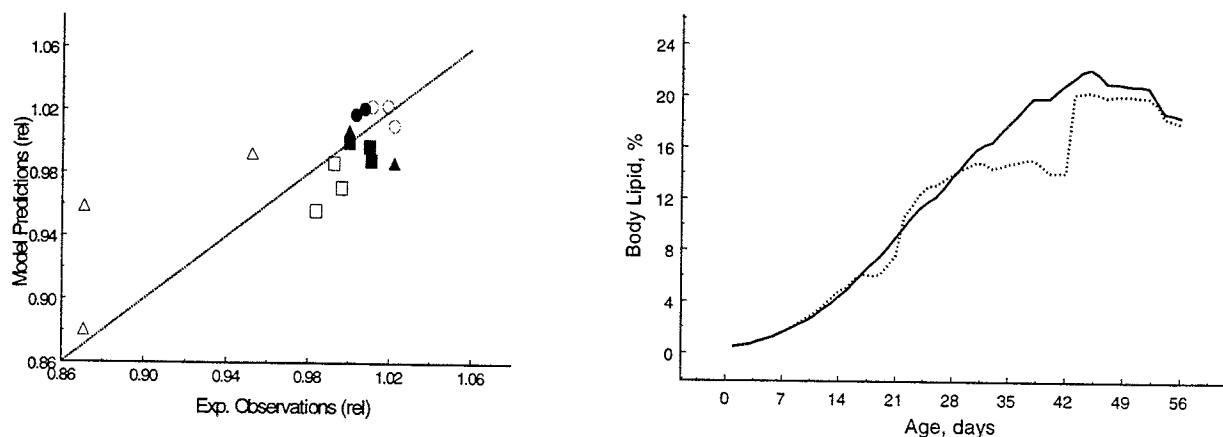


Figure 8. A feeding programme experiment (Saleh *et al.*, 1997). The left-hand figure shows the correlation between experimental observations and model predictions for body-weight (●), FCR (■), abdominal fat (▲), closed symbols refer to time of feeding the starter feed, open symbols time of feeding finisher feed. All data were scaled to a single treatment to facilitate presentation of the different responses. The right-hand figure shows the development of body lipid in model runs which emulate two of the treatments used.

APPLICATION AND VALIDATION TESTING OF A BROILER GROWTH MODEL AS A STRATEGIC PLANNING TOOL FOR LIVE PRODUCTION MANAGEMENT

B.I. FANCHER

Summary

Broiler production managers are increasingly challenged with ensuring that upon arrival at the processing plant, body weights of broiler chickens are within a specific range due to rigid requirements of the growing food service market sector. Animal growth models are generally regarded as tools for design of feeding programs, but their inherent ability to predict daily growth rate can be exploited to address scheduling of broiler harvest.

A software application derived from OmniPro®, a computerized broiler growth model linked to an economic optimizer, was developed for the above purpose and tested under commercial conditions. The application considers primary inputs of feed energy density, age at which diets are changed, environmental temperatures and live flock sample weights. The conducted test involved a comparison of predicted versus actual bodyweights under commercial conditions with and without the use of live flock sample weight inputs. Compared to common industry practices for predicting broiler harvest weight, the tested application offers substantial improvements in both accuracy (92% improvement) and variation (50% improvement) of prediction of marketing weight of broilers when utilizing live flock sample weight inputs.

I. INTRODUCTION

The path to profitability in animal production involves carefully choosing products made by comparing marketing prices and potential demand with expected production costs and capacity. As the enterprise proceeds with selection of target end products, the nutritionist is concomitantly challenged to design feeding programs which not only promote normal and healthy animal growth but also maximize net income by meeting marketing objectives and considering changing economic conditions. While the challenge is somewhat daunting, the nutritionist can exploit available growth modeling technology, particularly when linked to an economic decision making optimizer, to meet the challenge (Fancher 1996a,1996b,1997).

Animal growth models have generally been portrayed as tools useful for designing feeding programs to meet certain objectives. While currently available broiler growth models can be utilized to design the most profitable feeding programs, they can potentially be exploited by management for live production concerns. The inherent ability of a broiler model to predict daily growth is of value to management who must ensure that harvest of broilers occurs within the proper marketing weight range. The ability to harvest broilers of a particular size range is of increasing importance due to the growing food service market sector. In 1995, 30.3% of the total U.S.A. broiler production entered the food service market sector (National Broiler Council, 1995). Much of this market sector is concerned with fast food meat cuts having rigid size requirements. If an enterprise has a significant portion of its total volume tied to contractual agreements with fast food clients and flocks are harvested at the improper size, substantial economic losses will likely occur.

Omnus, Inc., 3000 Highway 40/61, O'Fallon, MO 63366.

Moreover, it is highly probable that birds harvested at an undesirable size will enter the "spot" market at a discounted price (i.e. a quick search for a buyer for the product due to limited storage capacity). Clearly, a model that accurately predicts broiler growth can be exploited to minimize the likelihood of harvesting birds at an undesirable size. This report describes the application of a broiler growth model for strategic planning of live bird production and testing to validate its effectiveness using actual broiler industry data.

II. METHODS

A software derivative of OmniPro, a computerized broiler growth model working in concert with an economic optimizer, was prepared specifically to meet requirements for use in scheduling harvest of broilers. The Hurwitz broiler model is the predecessor of OmniPro *per se* and has been described elsewhere (Hurwitz *et al.*, 1978,1980; Talpaz *et al.*, 1986,1988,1991). Harlow and Ivey (1994) performed a statistical evaluation demonstrating the precision, accuracy and commercial benefits of the Hurwitz broiler model. The process of validating the effectiveness of OmniPro in designing feeding programs to maximize net income using actual broiler industry data has been previously described (Fancher, 1996a,b).

One of the top ten broiler companies collaborated in validation testing of the modified OmniPro application's ability to predict broiler weights upon arrival at the processing plant. The primary inputs required by the application were in-house weekly average temperatures, dietary energy levels for all feeds, age at which diets are changed, bird gender, and the particular location's historical broiler growth performance. The latter was required in order to calibrate growth curves by sex status of birds. Estimates of farm to processing plant transport (i.e. "shrink") and feed withdrawal-related bodyweight losses were included in the application design. The software was also designed to consider live flock sample weights, if available, as means of further modification of the reference growth curve to reflect the respective flock's observed growth behavior.

Live flock sample weights were taken at 27-28 days of age from 18 different farms growing broilers of mixed sex. The results from these 18 farms represented a total of 790,551 processed broilers. Live flock bodyweight samples were taken by herding 100-150 birds into a wire enclosure and then weighing each captured bird individually on electronic scales. Field service technicians employed by the collaborating broiler company performed sampling. The sex status of each bird weighed was identified and matched up with the respective recorded liveweight. The mixed sex average was subsequently calculated as average of the mean male weight and mean female weight.

Prior to using the application for predictions, the mixed sex growth curve of the application was calibrated using the overall average growth rate (43.8 g/d growth; average weight 1792 g; average age 40.95 d), feed energy levels (diet 1, 13.47 MJ ME/kg; diet 2, 13.57 MJ ME/kg; diet 3, 13.69 MJ ME/kg), ages at diet change (diet 1, 0-16d; diet 2, 17-35d; diet 3, 36d-end) and in-house weekly average temperatures (0-7d, 31.1°C; 8-14d, 29.4°C; 15-21d, 27.8°C; 22-28d, 25.6°C; 29-35d, 24.4°C; 36-42d, 22.8°C) existing in the collaborating location during the period of live bodyweight data collection. Thereafter, based on inputs provided to the application (i.e. with or without live flock sample weights), predictions were made and compared with actual final flock weights as determined by the collaborating company.

III. RESULTS AND DISCUSSION

The first set of predictions for all 18 flocks made by the application were conducted without any inputs of live flock sample weights. Results using this initial approach appear in Table 1. The difference in predicted versus actual broiler weights at the processing plant ranged from -8.96 to 8.33%. The overall average difference was -1.11%. On average, therefore, the predictions appeared acceptable. More critical, however, is variation in accuracy of prediction since the broiler producer seeks to prevent any individual flock from arriving at the processing plant with weights outside of the tolerable range. In this sample population, the standard deviation (SD) of the average percent difference was 4.85.

Table 1. Omnis broiler growth model performance using no live flock sample weights prior to predicting processing plant broiler weights at 39-42 days.

Farm number	Broiler Cross	Termination age (days)	Final body weight		Difference (predicted vs actual) (%)
			Predicted (g)	Actual (g)	
1	A	40	1751	1807	-3.11
2	M	39	1707	1875	-8.96
3	B	40	1751	1784	-1.87
4	B	40	1751	1784	-1.87
5	B	40	1751	1757	-0.35
6	B	40	1751	1784	-1.87
7	C	39	1707	1816	-6.00
8	A	39	1707	1793	-4.81
9	A	39	1707	1757	-2.84
10	D	40	1751	1825	-4.07
11	B	40	1751	1616	8.33
12	B	40	1751	1657	5.65
13	B	40	1751	1821	-3.83
14	B	40	1751	1834	-4.54
15	B	41	1795	1821	-1.43
16	B	40	1751	1625	7.72
17	A	39	1707	1743	-2.08
18	B	41	1795	1693	5.97
Mean					-1.11
SD					4.85

Feedback reaction solicited from broiler companies indicates that desirable values for average percent difference and standard deviation are 0.00 and ≤ 1.25 , respectively when using a reference point of a 1770 g broiler target. However, it should be noted that the author is aware of no current bird harvest scheduling practices in the U.S. broiler industry, based on numerous evaluations of broiler company data, which meet their tolerance range. For example, one year of final bodyweight data from the collaborating complex (representing 49,927,039 broilers) were analyzed to determine the accuracy and variation of predictions for broiler body weights arriving at the processing plant when utilizing their in-house method. When comparing the predicted to actual weights, the average difference was -1.40% with a STD of 4.89. The commonly used method predicts harvest weight by estimating the total

amount of feed consumed by the flock to a tentative harvest date, dividing this number by the previous week's average feed conversion, and then dividing by the projected number of birds to be harvested. To allow reasonable estimates of total flock feed consumption and bird count, this prediction method is typically performed \leq one week in advance of tentative slaughter, which leaves minimal planning time. An alternate method used by the collaborator, similar to other U.S. broiler companies, for farms with a historical problem of erroneous bird size involves weighing a sample of birds on the farm two days prior to harvest, again providing minimal planning time. The overall performance using the computer application, without any live flock sample weight inputs, was comparable to the collaborator's in-house method in terms of accuracy and variation.

Table 2. Omnis broiler growth model performance using 27-28 day bodyweight sample data inputs to predict processing plant broiler weights at 39-42 days.

Farm number	Broiler Cross	Sample age (days)	Mean sample weight (g)	Termination age (days)	Final body weight		Difference (predicted vs actual) (%)
					Predicted (g)	Actual (g)	
1	A	27	1067	40	1925	1807	1.01
2	M	28	1162	39	1793	1875	-4.36
3	B	27	1053	40	1807	1784	1.27
4	B	28	1058	40	1743	1784	-2.29
5	B	28	1108	40	1798	1757	2.33
6	B	28	1094	40	1784	1784	0.00
7	C	28	1112	39	1739	1816	-4.25
8	A	28	1203	39	1843	1793	2.78
9	A	28	1162	39	1793	1757	2.07
10	D	27	999	40	1743	1825	-4.48
11	B	28	976	40	1648	1616	1.97
12	B	28	985	40	1657	1657	0.00
13	B	27	1071	40	1830	1821	0.50
14	B	27	1058	40	1811	1834	-1.24
15	B	28	1062	41	1811	1821	-0.50
16	B	28	985	40	1647	1625	1.96
17	A	28	1162	39	1793	1743	2.86
18	B	28	940	41	1666	1693	-1.61
Mean							-0.11
SD							2.46

The second set of predictions for all 18 flocks made by the application were performed using 27-28d live flock sample weight inputs. Results using this approach are provided in Table 2. The difference in predicted versus actual broiler weights at the processing plant ranged from -4.48 to 2.86%. Overall, the average difference was -0.11% with a STD of 2.46. Compared to the first approach, the use of live flock sample weights to modify the reference growth curve improved prediction accuracy and reduced variation. Assuming the sample population utilized here was representative of the collaborating broiler complex, the second approach would be expected to meet the U.S. broiler industry's requirements approximately 68% of the time. While still falling short of the desirable and

challenging requirement of the U.S.A. broiler industry, this approach is substantially superior to the common industry practices for predicting harvested broiler weights. In addition, use of the application provides a much longer planning horizon and requires less labor. The application described here predicts broiler weights on a daily basis. In practice, broilers are processed at differing times on a given day. Modification of the application to allow bodyweight predictions at intervals of time < one day would logically improve their accuracy since broilers can gain approximately 64g/d from 38-40 d of age.

The growth performance of broilers is obviously influenced by other factors in addition to temperature, feed energy density, and age of changing diets. Such factors are breed (which while documented here was not specifically calibrated for), age of parent flock of breeder hens, farm management practices *per se*, equipment used in the growing facility, feed additives, stocking density and numerous other factors. Builders of this sort of technology must face the challenges of determining, in the scheme of things, what matters and what does not - a time consuming and often tedious process. A discussion of problems associated with developing poultry growth models alone is provided by Emmans (1995). It is reasonable to expect that the ability of an application to predict broiler growth, as described here, can only improve when it is designed to account for factors identified as significant sources of variation affecting broiler growth. With appropriate design modifications, it therefore seems probable that broiler growth models can be exploited to solve chronic live production strategic planning problems such as scheduling the harvesting of broilers.

The acceptance of computerized models in the agricultural business sector is currently at a very early stage. However, currently available applications and those which will soon enter the marketplace present significant opportunities for incremental improvement and it is a mistake, in the author's opinion, to ignore their current value. Skellam (1973) offered sage advice when he stated, "Roughly speaking, a model is a peculiar blend of fact and fantasy, of truth, half-truth and falsehood. In some ways a model may be reliable, in other ways only helpful and at times and in some respects thoroughly misleading. The fashionable dogma that hypothetical schemes can be tested in their totality in some absolute sense is hardly conducive to creative thinking. It is, indeed, just as great a mistake to take the imperfections of our models too seriously as it is to ignore them altogether."

IV. CONCLUSION

Currently available broiler growth models have demonstrated practical usefulness in designing feeding programs to meet certain profit objectives; however, they can also be exploited to address management problems in the live broiler production sector. A computerized application derived from OmniPro is described which predicts broiler weights upon arrival at the processing plant after considering in-house temperatures, dietary energy density, age of changing diets, and live flock sample weight inputs. The application also allows for growth curve calibration. Compared to common broiler industry practices in the U.S. for scheduling the harvest of broilers, the application substantially improves accuracy and reduces variation in predicting broiler weight at harvest. To achieve the desirable and challenging requirements of the broiler industry for software performance in predicting broiler bodyweights, it appears likely that the software must account for other sources of variation affecting broiler growth beyond those already mentioned.

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APPLICATION OF COMPUTER SIMULATION MODELS IN INTENSIVE ANIMAL PRODUCTION

J.L. BLACK

Summary

Computer simulation models can be of enormous value for improving the profitability of intensive livestock enterprises. Simulation models provide the only realistic method for applying the bulk of the relevant information to the solution of many management problems. However, before models are likely to be adopted widely by industry, they must represent an appropriate level of biology, the predictions must be accurate, the monetary value of management changes must be predicted and they must be easy to use and interpret by non-scientific experts. Several examples of the areas where models can assist the management of pigs are given using the AUSPIG model.

I. INTRODUCTION

The primary objective of commercial poultry production is to maximise profit without compromising the welfare of either the birds or the staff. The net financial return from an enterprise depends on the interaction between many factors including those that affect the efficiency of production such as the strain and sex of bird, the diet composition and its form, the climatic environment, stocking arrangements and the prevalence and severity of disease. Other factors not related directly to the efficiency of production, including the premium paid for products with different specifications, the relative price structure of feed and products, the availability and cost of capital, labour, parent-stock and other resources, are important also in determining final enterprise profit.

Although there has been a great deal of research into many of the factors known to affect the efficiency of poultry production, the complexity of the interactions between them makes it virtually impossible for the human mind to assess accurately the consequences of alternative management strategies on either the efficiency of production or the long-term profitability of an enterprise. The human mind is great for inventing concepts and hypotheses about how systems operate and the factors that control them, but it is poor at following through time the consequences of changes in events. However, through the transformation of concepts and knowledge into mathematical algorithms and their integration into computer programs using simulation modelling techniques, the vast store of information available can be applied directly to improving the management, animal welfare and profitability of animal enterprises.

The value of animal simulation models has been demonstrated clearly through application of the AUSPIG decision support software (Black *et al.*, 1986; Davies *et al.*, 1993) in the pig industry. When introducing AUSPIG into different countries, it became apparent that the most successful producers in each country adopted practices that were then emulated by other producers to become "standards" for that country. However, these standard practices varied considerably between countries and many were shown, using AUSPIG, not to be either biologically or economically optimal. Enterprise profitability in several countries was increased by up to 30% through the adoption of new management practices which differed substantially between the countries.

The purpose of this paper is to: (i) define the minimum specifications for simulation models designed to improve the profitability of intensive livestock industries; (ii) provide examples of how AUSPIG has assisted the pig industry to improve profitability and; (iii) outline the measurements and information that must be obtained from animal enterprises to allow proper application of models.

II. SCOPE OF MODELS

Experience with the application of AUSPIG has shown that there are three essential specifications for simulation models if they are to be adopted widely by industry. The models must represent an appropriate level of biological detail, they must be accurate and they must predict the monetary value of changes made to management of an enterprise.

Models can be constructed at a range of levels which represent the system to be simulated in different degrees of detail. France and Thornley (1984) proposed a hierarchy for animal systems which ranged from the enterprise level to the flock, to the individual animal, to physiological and productive functions represented by organs, to metabolic functions represented by cells and organelles and finally to biochemical reactions and fluxes represented by macromolecules. It is essential that a model is constructed at some level below the one for which accurate predictions are required. For example, if a pig or poultry model is required to predict accurately under most circumstances changes in the efficiency of feed use, growth rate and body composition, it must be constructed at least down to the metabolic functions level and predict the utilisation of nutrient classes for various body functions.

Accuracy in prediction comes first through the appropriate mathematical representation of the perceived mechanisms controlling the system. Models that rely mainly on polynomial equations fitted to data without consideration of the underlying mechanisms generally fail when applied outside the range of the original data. Secondly, the models must be extensively tested and evaluated before they are introduced into the industry.

Mechanistic models that predict accurately animal performance are, by themselves, unlikely to be adopted widely by industry. Although such models are ideal for establishing research priorities, they are likely to be used for managing commercial enterprises by only a few specialist advisers. The animal simulation models must be integrated with other modules that address the major components of an enterprise that determine its profitability. It is essential also that the resulting software product is easy to use and interpret by industry personnel. For example, a model predicting amino acid and energy utilisation for intensive livestock should be integrated with several other modules including: (i) a least-cost diet formulation program that takes animal requirement specifications from the biological model; (ii) a linear program that maximises profitability by optimising the use of capital, labour, shed space and other resources in relation to the price paid for product by different buyers and the performance characteristics of the animals; (iii) data bases for commonly used information such as dietary ingredients, climate files, animal genotype parameters and specifications for frequently used graphs and tables and; (iv) an interface that is easy to use and model outputs that are readily interpreted. An Expert System that interprets predictions from the simulation module by identifying reasons for biological inefficiencies and recommends management strategies that should improve the efficiency of production and enterprise profitability is valuable also for assisting uptake of the software by industry. An Expert System has been incorporated into AUSPIG (Menzies *et al.*, 1988).

III. TESTING AND VALIDATION OF MODELS

Models that are to be accepted by industry must be thoroughly tested and evaluated before release. Testing and evaluation of models is an extremely important but difficult process. Several models constructed for the animal industries have failed commercially because inadequate attention was paid to the step of ensuring accuracy of prediction. The term testing is generally taken to mean that the model is mathematically, numerically and logically correct, that is, it is free from 'bugs'. Model testing is an objective process and can be conducted within a formal framework. There are several quality assurance procedures that should be adopted by software writers to reduce the chances of making errors and to increase the opportunity for locating those that have occurred.

Model evaluation, or validation as it is sometimes termed, is not such an objective process and is often difficult to accomplish. Model evaluation is concerned with establishing the "appropriateness" of a model for predicting outcomes within the system described by the model. A biological model is always only an approximate representation of a real system and rarely will it describe accurately the behaviour of a system under all circumstances. The aim of model evaluation is to determine the usefulness of the model within the limits of the scope for which it was intended. Models cannot be validated, they can be only invalidated. Model evaluation is a long and ongoing process. The fact that a model predicts accurately under one set of circumstances does not mean that it will do the same under other circumstances. The three main steps in model evaluation are: (i) checking the general behaviour of the model to ensure that the overall patterns of responses predicted are similar to those that would be expected by an expert in the field; (ii) undertaking sensitivity analyses to identify the equation components to which important predictions by the model are most sensitive and therefore which need to be known with most confidence and; (iii) making direct comparisons of model predictions with experimental results. Statistical procedures can be used to assess the accuracy of model predictions when compared with experimental observations. However, for such assessments to be valid, it is essential to assume that the experimental animals and the simulated 'animals' are from the same population (Black, 1995).

IV. EXAMPLES OF MODEL APPLICATIONS

Following are several examples where the AUSPIG decision support system has been used in the pig industry to help change management practices and improve enterprise profitability. Other examples, including the effects of protein content of the diet, restricted feeding, stocking density and ambient temperature, are given by Bradley (1994), de Lange and Schreurs (1995), Edwards (1997) and Smits (1997).

(a) Optimal use of dietary protein and free amino acids to reduce nitrogen loss in effluent

Traditionally, pig breeding companies have recommended the use of diets with excessively high protein contents to ensure that all animals from their breeding stock have the opportunity to express potential rates of body protein deposition. Conversely, the disposal of effluent from intensive livestock enterprises is becoming a major concern for environmental monitoring agencies and all new piggeries in Australia are likely soon to be licensed for maximum rates of nitrogen and phosphorus release. Computer models like AUSPIG are ideal for determining the consequences of reducing the protein content of the diet and improving the balance of amino acids within the protein on pig performance, profitability and nitrogen output in effluent.

Following is an example where entire male pigs of a specified genotype were fed a traditional high protein diet (Diet A) throughout the growth period from 50 to 100 kg live weight. AUSPIG was used first to predict the performance and profitability of pigs fed this diet and to estimate the amount of nitrogen lost to the environment. Predicted and observed performance of the pigs was similar and the net profit was predicted to be \$101.45 per pig (Table 1). However, the nitrogen content of the diet was excessive and 70% of the consumed nitrogen was lost to the environment. The high nitrogen loss occurred first because the protein content of the diet was above that required by the pigs weighing 50 kg; secondly, because the protein content of the diet remained unchanged throughout despite falling requirements of the pigs as they matured, and thirdly, because the amino acid balance of the protein was not close to that required by the animals.

Several other simulations were then conducted. Diet B was formulated using normal commercial ingredients to meet 103% of the requirements for amino acids of the average pig at 55 kg and this diet was again offered throughout the simulation. In treatment C, five diets were formulated using commercial ingredients to 103% of amino acid requirements for the pig at live weights of 50, 60, 70, 80, and 90 kg and each diet was offered at the corresponding live weight. In treatment D, five diets were again formulated to be fed at the same live weights as for treatment C, but total available nitrogen content of the diets was restricted to 120% of requirement to improve considerably the amino acid balance of the protein. This resulted in the substitution of some protein ingredients with free amino acids. It was not possible using commercial ingredients to reduce available nitrogen content of the diets much below 120% of requirement and meet all other constraints when formulating the diets. Isoleucine, tryptophan, valine, threonine, methionine and lysine were added to these diets. The cost of amino acids not commonly used in pig feeds was assumed to be \$ 8,000/t.

The results of the simulations given in Table 1 show an advantage of \$2.00/pig in profit and a 25% decrease in nitrogen excretion from treatment C when five diets which closely matched protein requirements of the animals were fed compared with the traditional feeding regime. However, the greatest reduction in nitrogen excretion resulted from replacement of some of the protein of conventional ingredients with free amino acids. Treatment D resulted in a 5% decline in profit relative to treatment C, but a further 40% reduction in nitrogen excretion.

This example illustrates that, compared with traditional diet and feeding regimes, an increase in profit and a substantial decrease in nitrogen loss to the environment would result from feeding several diets that meet more closely the requirements of the animals. However, the long-term profitability of reducing nitrogen excretion by improving the balance of amino acids in the protein offered to pigs will depend on the future price of free amino acids and the cost of nitrogen disposal.

(b) Effect of the digestible energy content of cereal grain on piggery profitability

Recent studies by Kopinski (1997) and van Barneveld (1997) have shown that the digestible energy content for pigs of cereal grains grown in Australia can vary by as much 3 MJ/kg for the same species depending on the cultivar and growing conditions. For example, values for wheat grain were found to range from 13.3 to 17.0 MJ/kg (van Barneveld, 1997). Kopinski (1997) has used AUSPIG to assess the consequences of a difference in digestible energy content of wheat grain of only 0.7 MJ/kg on the profitability of a 200 sow herd in Queensland. Table 2 shows the predicted effect on piggery profit of this 5% reduction in the digestible energy content of wheat assuming the diet was formulated without knowledge of the reduction. Such a situation would commonly occur in practice because of the lack of

reliable and rapid methods for estimating the digestible energy value of grains for pigs. The lower digestible energy content of the cereal grain in the diets for the piggery was predicted to result in no change in the growth performance of the pigs from birth to slaughter but a significant increase in the amount of feed eaten. Over the whole growth period an extra 70 g more feed was predicted to be used for every 1 kg of liveweight gain. The increase in feed eaten resulted in a reduction in profit of \$1.95/pig sold and this translated to a lowering of over \$7500 in the annual profit of the piggery. A similar analysis for wheat priced at \$240/tonne, as can occur during droughts, gave a difference in annual piggery profit of \$14,904.

Table 1. Predicted performance, profitability and nitrogen (N) excretion of male pigs grown from 50 to 100 kg when offered the series of diets described above (a).

	Observed Diet A	Predicted Diet A	Diet B	TreatmentC	Treatment D
Diet Protein (%)	21.2	21.8	18.6	18.5 18.2 17.8 17.2 16.6	15.3 15.0 14.5 13.5 11.8
DE (MJ/kg)	14.7	14.9	14.5	14.5	14.5
\$/tonne	271	271	263	264 261 258 253 247	298 294 292 293 310
Feed Intake (kg/d)	2.34	2.32	2.37	2.36	2.23
Live weight gain (g/d)	879	878	872	874	879
P2 back-fat (mm)	11.3	11.6	11.3	11.4	11.5
Feed gain	2.67	2.60	2.67	2.65	2.49
Gross return (\$/pig)		201.68	202.66	202.88	201.67
Feed costs (\$/pig)		35.23	35.62	34.40	37.91
Profit (\$/pig)		101.45	102.04	103.47	98.75
Total N intake (g)		4123	3653	3443	2530
(g/d)		73.7	64.1	60.4	45.2
Total N excretion (g)		2879	2378	2169	1275
(g/d)		51.4	41.7	38.1	22.8
N excretion (% intake)		70	65	63	50
(% Diet A)		100	83	75	44

(c) Cost of feed wastage

Results from studies by G.D. Hudson (unpublished) show that feed wastage can range from 2.5 to 40% of the feed offered to pigs depending on the physical form of the feed and the type and adjustment of the feeder. AUSPIG was used to assess the effect of feed wastage on the profitability of the 200 sow reference herd (Table 3). The reference herd was established using the mean values for growth and reproductive performance characteristics of Australian piggeries from 1995 PigStats (Meo and Cleary, 1996). Feed waste was altered only in the grower herd and was assumed to be unaltered for breeding sows and boars. Feed

wastage was changed from 0 to 5% of feed intake, but actual feed intake and performance was assumed not to be affected by feed wastage.

Table 2. A comparison of pig performance and financial returns when the actual digestible energy (DE) content of wheat is either equal to or 5% less than the value used to formulate the diets for the piggery.

Variable	Actual DE equals formulated DE	Actual DE is 5% less than formulated DE
Assumed DE of wheat at formulation (MJ/kg)	14.3	14.3
Actual DE of wheat (MJ/kg)	14.3	13.6
Wheat cost (\$/tonne)	190	190
Live weight gain from birth (g/d)	555	555
Feed:gain from birth	2.62	2.69
Profit (\$/pig)	9.66	7.71
Profit (\$/sow/y)	187	149
Profit for 200 sow piggery (\$/y)	37386	29818

Table 3. Predicted effect of feed wastage on profitability of a 200 sow piggery.

Feed Waste (%)	Feed Costs (\$/y)	Profit (\$/y)	Total feed (t/y)	Feed/sow/year
0	348373	204022	1109.5	5.545
4	358427	193827	1141.3	5.705
8	368672	183724	1172.9	5.865
12	378867	173529	1204.7	6.024
15	386409	165987	1228.3	6.142

The annual income and non-feed costs were unaffected by feed wastage and predicted to be \$789161 and \$236765, respectively. Profitability of the 200 sow reference piggery was shown to increase by \$2548 for every 1% decline in feed wastage. This represents \$12.70/sow/year for each 1% of feed intake wasted.

The average feed wastage of Australian pig herds is hard to judge. The average feed use/sow/year in PigStats95 is 5.757, which is equivalent to a feed wastage of 5.3% when compared with the values given in Table 3. Commonly, values for feed waste of between 10 and 15% are required in AUSPIG to predict observed herd feed conversion ratios when simulating Australian piggeries. However, in one recent example, feed wastage of almost 40% was required to simulate accurately feed usage by the whole herd. In the above example, such a feed waste would cost the reference piggery over \$100000/year and reduce profit to 33% of the value if feed waste was controlled to only 4% of feed intake.

(d) Carcass price needed when pigs are sold at sub-optimal weights

As a final example of the way models can help improve profitability of farms, AUSPIG is used to determine the price that would be needed to a producer if the buyer requests pigs at lighter weights than is optimal under the current selling system. The example uses a standard buyer's price matrix in which the price/kg carcass varies with carcass weight and back-fat thickness. For the particular example simulated, the profit was maximised when

10% of pigs were sold at a carcass weight of 70kg, 75% at 75kg and 15% at 80kg. The predicted effect on price required per kg carcass, if profit was to remain unchanged, of selling all carcasses at either 50 or 60kg was determined under two circumstances: (i) assuming that the number of sows could not be increased or (ii) assuming that total floor space was limiting but could be redistributed between different stock classes (Table 4).

Table 4. Predicted effect of selling pigs at sub-optimal weights on annual profit relative to optimal selling weights and on the price needed/kg carcass to restore profit.

Simulation	Relative number of sows	Relative profit	Relative number of pigs sold	Relative loss (% profit)	Loss (\$/pig sold)	Price increase needed to restore profit (\$/kg carcass)
Normal optimal selling	100	100	100	-	-	-
50 kg carcasses						
Sows limited	100	32	101	68	35.69	0.71
Floor space limited	131	53	132	45	18.14	0.36
60 kg carcasses						
Sows limited	100	64	100	36	18.80	0.31
Floor space limited	116	81	116	19	8.82	0.15

The simulation shows that the price would have to be increased above the current price schedule by between 0.15 and 0.71 \$/kg carcass if the profit from the piggery were to be maintained. Such information is essential for both buyers and producers if acceptable price matrices are to be negotiated.

V. CONCLUSIONS

Accurate, mechanistic computer simulation models that predict the effects of genotype, diet, environment, social interaction and disease on the efficiency of nutrient use and body composition can be of enormous value for improving the management of intensive animal enterprises. The value of these models is enhanced substantially if they also predict the monetary effects of management changes. However, to apply models satisfactorily to commercial operations, considerable effort must be put into recording animal performance, feed consumption, diet composition, climatic conditions and other aspects of day-to-day operations. When this is done, the rewards in terms of increased profitability can be highly rewarding. Examples of the use of the AUSPIG software around the world demonstrate that simulation models may be the only way of satisfactorily integrating the mass of information available in a way that enables it to be focused onto real farm issues and increase profitability.

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AMINO ACID NUTRITION FOR POULTRY IN HOT CONDITIONS

P.A. GERAERT

Summary

The rapid development of poultry production in hot climates places great emphasis on finding solutions to alleviate the depressions in growth and laying performance which occur under these conditions. In broilers, the growth reduction, which is only partly explained by decreased feed intake, is also accompanied by enhanced fatness and reduced protein deposition. Drastic changes in protein and amino acid metabolisms occur under heat exposure. It appears that means which reduce fat deposition, such as higher amino acid intake and a better balanced supply, could reduce the consequences of heat distress. Complementary means should also be developed to help the birds withstand such drastic conditions.

I. INTRODUCTION

Ambient temperature is an important determinant of bird performance. Hot conditions, particularly, are causing increasing concern due to the rapid development of the poultry industry in hot climates and to the reduced performance of poultry recorded during summer months in temperate climates. Hot conditions may correspond to either chronic heat exposure (several weeks at high ambient temperature) or acute heat exposure (heat stress) (a few hours at very high temperature). Responses to these two different conditions do not evoke the same mechanisms. While heat stress might be alleviated through physical means (increased ventilation rate, use of cooling devices) to decrease peak temperature, the prolonged exposure might be tolerated through nutritional adjustments.

Birds, like mammals, are homeotherms, and are thus able to maintain a near-constant body temperature. To achieve a constant body temperature, heat produced by metabolism must equal heat loss. In birds, heat losses are limited by feathering which is an efficient insulation and by the absence of sweat glands. The main consequence of heat exposure is a reduction in feed intake in order to reduce metabolic heat production. In broilers, this reduction is approximately 1.5 to 2.5 % per °C increase in ambient temperature above 20°C and increasing with age and temperature (Howlider and Rose, 1987) and in layers reaches 2.5 to 4 g per °C (Nys, 1995). This reduced feed consumption leads to growth depression and lower egg production. However, the reduction in growth or in egg production is often greater than the reduction in feed intake, resulting in a lower feed efficiency (Howlider and Rose, 1987; Picard *et al.*, 1993).

Nutritional knowledge gained from the responses of birds under temperate conditions, accumulated over many years, has led to the belief that increasing dietary protein concentration by its resulting increase in heat increment would further impair performance under hot conditions. Thus the most common nutritional solutions proposed to help birds cope with hot conditions are, a) decreasing the dietary protein level, and b) increasing the fat content, thus reducing the protein : energy ratio to lower heat increment. Whilst such a solution seemed promising, the observed results have generally not been sufficiently promoted to have wide-spread acceptance.

Rhône-Poulenc Animal Nutrition, 42 Av A. Briand BP 100 92164 Antony Cedex, France.

A statement often made by poultry nutritionists is that temperature does not change requirements for protein (Daghir, 1995). As in the same manner as under thermoneutral conditions, rebalancing the dietary amino acid profile and thus allowing a decrease in total protein concentration through the use of synthetic amino acids has often been proposed to counteract the effect of chronic heat exposure. Unfortunately, such a strategy does not appear to be any more successful than reducing the protein to energy ratio.

This paper discusses: the discrepancy between nutritional theory, mainly acquired in thermoneutral conditions, and practical performance results obtained in hot climates; the profound modifications induced by heat exposure at the metabolic level; the potential role of choice-feeding experiments in providing a preferred nutritional solution by the birds; strategies based on high dietary protein or balanced amino acid supply; and advantages of genetically leaner birds in relation to the protein content of the diet.

II. REDUCED FEED INTAKE DOES NOT EXPLAIN THE WHOLE HEAT EFFECT

Using pair-feeding techniques, Geraert *et al.* (1996) demonstrated that about half of the reduction in growth due to chronic heat exposure was not related to feed intake, and thus could have other origins. Moreover, enhanced fatness has been observed in heat-exposed chickens despite their lower feed intake. Howlinder and Rose (1987) found an increase of 0.8 and 1.6 % in body lipid content and in abdominal fat proportion respectively per degree rise in temperature between 21 and 29°C. This increased fatness has been further analyzed by Aïn Baziz *et al.* (1996) who found that abdominal, subcutaneous and intermuscular fat deposits were enhanced in hot conditions (32°C) : by 15, 21 and 22 % compared to the control group (22°C), and by 58, 64 and 33 % compared to the control pair-fed exposed birds (Table 1). Moreover, in heat-exposed chickens, saturated fatty acid proportions, particularly palmitic acid (C16:0) were increased and conversely, unsaturated fatty acid percentages were decreased especially oleic and linoleic acids. Consequently, heat exposure significantly decreased the unsaturated to saturated fatty acid ratio in abdominal and subcutaneous fat tissues. These significant changes in lipid deposition suggest profound modifications in metabolism.

Table 1. Effect of heat exposure (4 to 7 weeks of age) on lipid deposition in *ad libitum* heat-exposed (32AL), *ad libitum* control-exposed (22AL) and pair-fed control-exposed (22PF) 7-wk old male broilers (after Aïn Baziz *et al.*, 1996).

	22AL	22PF	32AL
Final Body Weight (g)	2372a	1905b	1660c
Feed Conversion Ratio (g :g)	2.22a	2.11a	2.73b
Abdominal Fat ¹	2.85a	1.86b	3.28a
Subcutaneous Fat ²	5.80b	3.94c	7.01a
Intermuscular Fat ²	2.90b	2.59b	3.53a
Intramuscular Fat ³			
Breast	1.50ab	1.32b	1.67a
Leg	4.04ab	3.80b	4.50a

¹ in g/100g body weight; ² in g/100g leg weight; ³ in g lipid/100 g tissue
Means in a row not followed by the same letter differ significantly (P<0.05).

Protein retention also appeared to be reduced in heat-exposed broilers (Bonnet *et al.*, 1997; Tesseraud *et al.*, 1997) (Table 2). Even when taking into account the reduction in feed intake, nitrogen retention is decreased by up to 30 % (Tesseraud *et al.*, 1997). Geraert *et al.* (1996) observed that after two weeks at 32°C, protein gain decreased by 54 % and protein efficiency by 46 %.

Table 2 Effect of chronic heat exposure (4 to 6 weeks of age) on nitrogen (N) ingested, excreted and retained in *ad libitum* heat-exposed (32AL), *ad libitum* control-exposed (22AL) and pair-fed control-exposed (22PF) 7-wk old male broilers (after Bonnet *et al.*, 1997).

	Diet 1 (20 % CP)			Diet 2 (18.8 % CP)		
	22AL	22PF	32AL	22AL	22PF	32AL
N intake (g)	16.7a	10.4b	9.8b	14.8a	9.9b	9.4b
N excreted (g)	8.2a	5.0b	5.5b	7.4a	5.0b	5.6b
N retention (%)	51.1a	51.9a	43.2b	49.9a	49.9a	40.4b

Means in a row, within a diet, not followed with the same letter differ significantly ($P < 0.05$)

III. CHRONIC HEAT AFFECTS PROTEIN METABOLISM DEPENDING ON MUSCLES

In recent years, protein metabolism has received much more attention compared to previously. Reduction in protein deposition depends on the muscle type. Indeed, while most authors have recorded a reduction in the range 10 to 15 % in the pectoral muscle in proportion to body weight (Tesseraud *et al.*, 1997; Aïn Baziz *et al.*, 1996), muscle proportion in the legs often appears enhanced, suggesting differential effects according to muscle metabolism and main energy substrate. While the pectoral muscle is mainly glycolytic and uses glycogen as its main energy source, the sartorius and gastrocnemius muscles from the thigh and drumsticks are more oxidative and use fatty acids as their main source of energy.

Using the large dose technique (flooding dose of 3H-Phe), Temim *et al.* (1997) demonstrated that chronic heat exposure reduced protein synthesis and breakdown, thus reducing protein turn-over. This was associated with significant decreases in the capacity for protein synthesis and in the translational efficacy. The reduced protein deposition observed under heat exposure was explained by a greater decrease in protein synthesis compared with protein breakdown.

IV. HEAT-EXPOSED BIRDS PREFER HIGH PROTEIN DIETS

When placed in a choice-feeding situation, broilers selected a greater proportion of a high compared with a low protein diet (Figure 1; Hruby, 1995). However, irrespective of ambient temperature, chickens consume less protein with increasing age. Recently, MacLeod and Dabutha (1997) presented similar results obtained in heat-exposed quail having the choice between 450 g and 100 g crude protein/kg diets. The high protein diet represented 62 and 36 % of the total feed intake of the birds under the hot and thermoneutral conditions respectively. However all previous studies have not shown the same results. This might relate to the degree of imbalance in the dietary amino acids in these specific experiments. The enhanced intake of the high protein diet might suggest an increased need for protein

under hot conditions either associated with the overall decreased food intake or a more specific need in order to maintain protein deposition and thus growth.

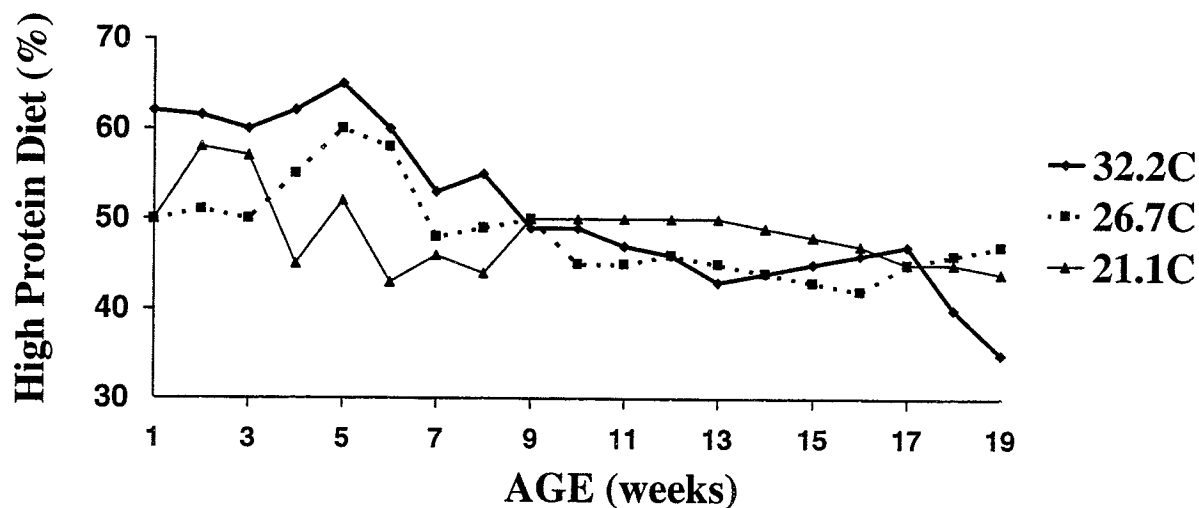


Figure 1. Effect of ambient temperature on choice-feeding between high (25% CP) or low (8.8% CP) protein isoenergetic diets in chickens from 1 to 19 weeks of age (after Hruby *et al.*, 1995).

V. IDEAL AMINO ACID BALANCE MIGHT DIFFER UNDER HEAT EXPOSURE

Due to its lower heat increment, fat supplementation has often been proposed as a means of enhancing feed intake at high temperature. However, the higher net energy content of fat counteracts the benefits in terms of energy intake and also increases fat deposition. Indeed, using a wide range of diet composition, 50 to 150 g/kg lipids and 2800 to 3300 kcal ME/kg, Aïn Baziz *et al.* (1990) could not find any gain in protein deposition under hot conditions but only changes in fat deposition. Surprisingly, increasing the dietary protein content from 170 to 300 g/kg did not result in enhanced heat increment as expressed by the slope of the regression between heat production and ME intake, suggesting different metabolic pathways under hot conditions. Recently, Padilha (1995) showed that enhancing the total protein content of the finisher diet from 15 to 25 % resulted in an increased weight gain under constant high temperature (32°C) while under thermoneutral conditions, weight gains of chickens plateaued beyond 20 %. Similarly, in layers a positive effect of high protein diets has been reported by Uzu (1989).

Is the positive effect of high protein diets due to increased needs for some amino acids? Dietary supplementation with common amino acids used in poultry nutrition such as methionine, lysine or threonine has not always shown significant improvements. Rose and Salah Uddin (1997) found a significant lysine balance x temperature interaction. The relative changes in growth rate were less affected by lysine to protein ratio at 30°C than at lower temperatures. The effect of temperature on lysine requirement could also depend on sex, being more important for females, as revealed by Han and Baker (1993). Moreover, Balnave and Oliva (1990) reported a lower need for methionine under constant than cycling hot temperatures (0.22 vs 0.26g methionine per MJ energy). Austic (1985) and Waldroup (1982), reviewing the literature, concluded that there was no evidence for an increased need in amino acids above 32°C. However blending protein sources in combination with synthetic amino

acids might help to restore performance under hot conditions. Recently, Alleman and Leclercq (1997) compared a standard protein diet (20 %) to a low protein diet (16 %) but rebalanced with synthetic amino acids. Lysine, sulphur amino acids and the other essential amino acids were added according to the latest requirements obtained under thermoneutral conditions (Leclercq *et al.*, 1997). High temperature reduced growth rate, feed efficiency, as well as breast meat proportion but increased fatness. Whereas under thermoneutral conditions, both diets gave same performance and breast meat deposition, under hot conditions, low-protein fed broilers exhibited lower weight gain and decreased breast meat deposition (Table 3).

Table 3. Ideal amino acid balance measured at thermoneutrality (22°C) and under hot (32°C) conditions (after Alleman and Leclercq, 1997).

Temperature	22°C		32°C	
	Protein (g/kg)	160	200	160
Live weight gain (g)	1783 ^c	1779 ^c	939 ^a	1118 ^b
Feed intake (g)	3256 ^c	3108 ^b	2279 ^a	2333 ^a
Feed conversion ration (g :g)	1.811 ^b	1.772 ^a	2.413 ^d	2.194 ^c
Abdominal fat (g/kg)	2.78 ^{ab}	2.20 ^a	3.77 ^c	3.24 ^b
Breast meat (g/kg)	14.7 ^c	15.4 ^c	12.1 ^a	13.5 ^b

Finally, an interesting strategy, based on enhancing the arginine to lysine ratio, was recently proposed by Brake and Balnave (1995). Arginine supplementation appeared to have a beneficial effect on viability under acute heat stress (Table 4). Metabolic needs for arginine would thus appear to be increased while at the same time its availability is reduced. Using the *in vitro* methodology with intestine fragments, Dibner *et al.* (quoted by Brake and Balnave) showed that whereas under temperate conditions, arginine and lysine uptakes were similar, under hot conditions arginine uptake by the intestine dropped significantly compared with lysine uptake. Such a discrepancy would create an imbalance under hot climates.

Table 4. Effect of arginine: lysine ratio on weight gain (W.G.), feed conversion ratio (FCR) and mortality in chickens exposed to 21 or 31°C and submitted at 43 days of age to a heat stress (35°C) (after Brake and Balnave, 1995).

T (°C)	Ratio	W.G.	FCR	Mortality	35°C-43d
21	1.05	1124	2.18	0	25
	1.15	1142	2.14	0	25
	1.25	1159	2.15	0	35
	1.35	1110	2.20	0	30
31	1.05	971	2.12	15	15
	1.15	980	2.13	5	10
	1.25	988	2.10	5	6
	1.35	1011	2.17	0	0

To reconsider the amino acid balance, it might be worthwhile to investigate the plasma amino acid profiles of heat-exposed and control-exposed broilers. Geraert *et al.* (1996) found significant decreases in all plasma amino acids except for aspartic and glutamic acids. Padilha (1995) demonstrated that supplying a high protein balanced diet could significantly decrease the difference between plasma amino acid profiles at both temperatures and thus greatly improve growth under hot conditions. Thus ideal amino acid balance differs under different ambient temperatures.

VII. IS THERE AN EFFECT OF METHIONINE SOURCE?

It is difficult to find constant and significant difference in bioefficacy between methionine (DLM) and methionine hydroxy analogue (HMB) under hot conditions. Rostagno and Barbosa (1995) compared the bioequivalency of DLM and HMB under hot conditions and found similar values compared with those obtained at thermoneutrality. The only trials showing slightly better performances, feed conversion ratio and nitrogen retention are those from Swick *et al.* (1990;1991) and were mainly published in Conference Proceedings. Balnave and Oliva (1990), Wiernusz and Teeter (1994) and Teeter *et al.* (1996) could not demonstrate any difference between methionine sources under hot constant or cyclic conditions.

The scientific basis to account for these differences in efficacy between both sources was first thought to be linked to divergence in absorption or transport mechanisms. Hot conditions have indeed been demonstrated to affect digestibility and intestinal absorption (Bonnet *et al.*, 1997; Mitchell and Carlisle, 1990). However, large discrepancies exist between *in vitro* methodologies to measure transport mechanisms. The two forms of methionine are not absorbed by the same mechanism. While DLM is mainly absorbed through a broad specificity energy and Na-dependent neutral amino acid B-type transporter, HMB uses a non-stereo specific H⁺-dependent intermediate affinity transport system (Maenz and Engel-Schaan, 1996a;1996b). However even in this area controversy still exists. While Dibner *et al.* (1992) and Knight *et al.* (1994) wrote that diffusion was very important for HMB, Maenz and colleagues could not demonstrate such a mechanism. Indeed diffusion could be less affected by heat-induced changes in physiology than active mechanisms.

Finally, as indicated by Dr Maenz, caution must be exercised when trying to extrapolate *in vivo* consequences from *in vitro* measurements. Direct measures of *in vivo* passage rate and clearance in the intestinal lumen have to be performed to further understand whether differences might exist between the two forms of methionine. However, there is no clear evidence based on performance to show differences between methionine forms.

VIII. SELECTION FOR INCREASED LEANNESS IMPROVES HEAT RESISTANCE

Recent studies have shown that genetically lean broilers are more resistant to hot conditions, showing enhanced weight gains and better feed and protein efficiencies than their fat line counterparts (Geraert *et al.*, 1993; Cahaner *et al.*, 1995) despite the fact that the lean birds showed a higher heat increment and increased feathering (Geraert *et al.*, 1993). Leenstra and Cahaner (1992) also reported that broilers selected for improved feed conversion (FC line) showed the best growth rate and the lowest body fat content in a hot climate. Lean and FC genotypes are characterized by a lower energetic efficiency, i.e. an increased heat production per MJ ingested, which might signify improved heat loss capacity. Moreover, the lean genotypes appeared more efficient in transforming high protein supply into weight gain and lean mass deposition under hot climatic conditions than fat ones (Table 5).

Table 5. Effect of genotype (selection on low, LF, or high, HF, abdominal fat proportion or selection on bodyweight gain, WI) and dietary crude protein content (high : 227 and low : 161 g/kg) on growth, feed conversion ratio (FCR) and carcass components of broilers reared at high ambient temperature between 4 and 8 weeks of age (Cahaner *et al.*, 1995).

Line	LF		WI		HF	
	High	Low	High	Low	High	Low
Dietary protein content						
Weight gain (g)	1198	1050	982	1057	1110	1034
FCR	2.60	2.80	3.02	2.99	2.84	2.95
Abdominal fat (% BW)	2.18	2.33	2.86	3.42	3.64	5.28
Breast meat (%BW)	16.5	14.9	14.8	14.2	14.4	13.8

IX. CONCLUSION

Nutrition would thus appear to be a potential means of alleviating some of the loss of performance under hot conditions. However a better knowledge of the effects of heat exposure on amino acid metabolism is first necessary to design adequate diets. Indeed, in broilers, the better resistance of lean genotypes suggests that protein metabolism might be the key factor to improving performance under hot conditions. Dietary protein or amino acid supplementations would be beneficial. However, whereas most of the knowledge about amino acid nutrition has been obtained under thermoneutral conditions, few studies have looked at the requirements under hot conditions. Ideal amino acid balance appears to depend on ambient temperature. Finally, feed digestibility, particularly protein and amino acid intestinal absorptions which were lowered under hot conditions, should also be improved under hot conditions to avoid further imbalances. In that respect, use of in-diet enzyme supplementation might reduce disturbances in feed transit, endogenous enzyme activities and absorption mechanisms.

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HIGH-TEMPERATURE NUTRITION OF LAYING HENS

D. BALNAVE

Summary

The major effect of high temperatures on the performance of laying hens is exerted through a reduced feed consumption although the effects of heat stress *per se* vary with different measures of output. Attempts to define nutritional requirements at high temperatures have concentrated on energy and protein (amino acids) because of their economic importance. The results of studies involving the self-selection feeding of separate energy-rich and protein-rich feeds balanced for other nutrients, or feeding complete diets containing increased concentrations of protein, indicate that protein intake rather than energy intake is the most important factor in maintaining optimum egg production at high temperatures. Fats and oils are important energy sources with low heat increment and, in addition, their linoleic acid content can be used to manipulate egg weight. Although plasma phosphorus levels are reduced at high temperatures, increases in dietary phosphorus concentrations adversely affect egg shell quality. However, egg shell quality is improved by dietary supplementation with sodium bicarbonate as long as hens have access to the bicarbonate during the period of egg shell formation. Dietary ascorbic acid supplementation has been shown to counteract the adverse effects of heat stress on liveability, egg production, egg weight and shell quality.

I. INTRODUCTION

High temperature stress impacts adversely on all aspects of laying hen performance, including increased mortality as well as reduced appetite, egg production, egg weight and egg shell quality. The primary effect is exerted through reduced feed consumption but the relative importance of reduced nutrient intakes and heat stress *per se* varies with different measures of performance (Smith, 1972; Emery *et al.*, 1984; Picard, 1985).

The effects of heat stress can be ameliorated in a number of ways. These include procedures which induce the bird to increase its intake of water and, thereby, maintain more efficient respiratory evaporative heat loss, as shown with broilers by Belay and Teeter (1993). The use of cyclical daily temperatures, in which the bird is able to dissipate heat at low temperatures during part of the 24 h cycle, is also useful although the mean temperature and the amplitude of the temperature cycle are important factors to consider (Harris *et al.*, 1974; De Andrade *et al.*, 1977; Deaton *et al.*, 1981). However, since reduced appetite is the major factor associated with reduced performance at high temperatures, efforts continue to be made to define more accurately the nutrient requirements of laying hens under heat stress conditions.

II. ENERGY AND PROTEIN

The economic importance of feed ingredients supplying energy and protein has meant that the majority of nutritional studies at high temperatures have centred on examining the requirements for these nutrients.

Department of Animal Science, University of Sydney, Werombi Road, Camden, NSW 2570.

It is normally assumed that a limitation in energy intake is the most important factor contributing to reduced laying performance at high temperatures (Smith, 1973; Picard, 1985). Data, such as those reported by Smith and Oliver (1972) and El Jack and Blum (1978) showed large reductions in metabolizable energy (ME) intakes at high temperatures which were associated with corresponding reductions in bodyweight, egg production and egg weight. Furthermore, El Jack and Blum (1978) concluded that the decrease in egg production was not related to a protein or amino acid deficiency since the daily protein intake remained above 15 g/d. In addition, improving protein intake by increasing the dietary protein concentration of complete, isoenergetic diets only partially overcame the adverse effects of high temperatures on egg output in short-term studies of a few weeks duration (Valencia *et al.*, 1980; Scott and Balnave, 1988a).

Although feeding high-nutrient density diets to laying hens at high temperatures has not proved to be a completely successful strategy there is evidence to indicate that substantial benefits can sometimes accrue from adjustments to dietary protein and energy concentrations. De Andrade *et al.* (1977), in a short-term study of 12 weeks duration, maintained similar egg production when hens were fed a high nutrient density diet containing 10 percent additional ME and 25 percent additional protein at constant 21°, constant 31° or fluctuating 26°-36°C temperatures. Lin *et al.* (1993) reported that at ambient temperatures of 17°-25°C protein and ME intakes were the major factors influencing egg production whereas at 29°-33°C the major factors were protein and total phosphorus intakes. Further support for the suggestion that protein rather than energy intake is the most crucial factor in maintaining optimum egg production at high temperatures has been derived from self-selection studies.

(a) Self-selection feeding

An attempt by Blake *et al.* (1984) to improve the performance of 34-week old laying hens at high temperatures by self-selection feeding using three diets high in energy, protein or calcium proved unsuccessful. The authors assumed the lack of response was due to an inability of the hens to satisfy daily nutrient requirements from three separate diets. However, it is more likely that the problem was associated with a failure to acclimatize the hens to self-selection feeding prior to the onset of lay. Therefore, a reduction in egg output during the time the hens were defining their selection requirements probably impacted on feed consumption which, in turn, reinforced the lower egg output. It is interesting to note that the lower rate of lay at 30°C with the self-selection hens was associated with a reduced protein, rather than ME, intake.

The importance of acclimatising hens to self-selection feeding prior to the onset of lay was clearly shown in a series of studies by Scott and Balnave (1988b, 1989). Hens were allowed to select from two feeds, these being an energy-rich and a protein-rich feed, balanced for other nutrients. These studies found that laying hens introduced to self-selection feeding about five weeks prior to sexual maturity were subsequently able to accurately select nutrients to maintain normal rates of egg production in a diurnally cycling 25°-35°C environment. These hens showed an increased preference for protein commencing about three weeks prior to sexual maturity and this elevated intake plateaued approximately six weeks after the production of the first egg. The nature of this response is shown in Figure 1, which is taken from a 20-week study by Balnave and Murtisari Abdoellah (1990). Even with the low feed intakes prevalent at high temperatures this procedure substantially improves protein intakes without any major changes in ME intake (Scott and Balnave, 1988b, 1989; Balnave and Murtisari Abdoellah, 1990). Egg mass output is improved by up to 10 percent and appears to be associated with the ability of the self-selection fed hens to gain bodyweight at high

temperatures and not to draw on body reserves to maintain egg production. Similar responses to self-selection feeding are observed at cool temperatures but the increased energy requirements make the protein intakes uneconomically high without any improvement in egg mass output (Scott and Balnave, 1988b). In a similar way Picard (1985) reported that, whereas allowing hens to self-select calcium from a separate feeder improved egg output at 33°C, no such response was obtained at 20°C where nutrient intakes were excessive.

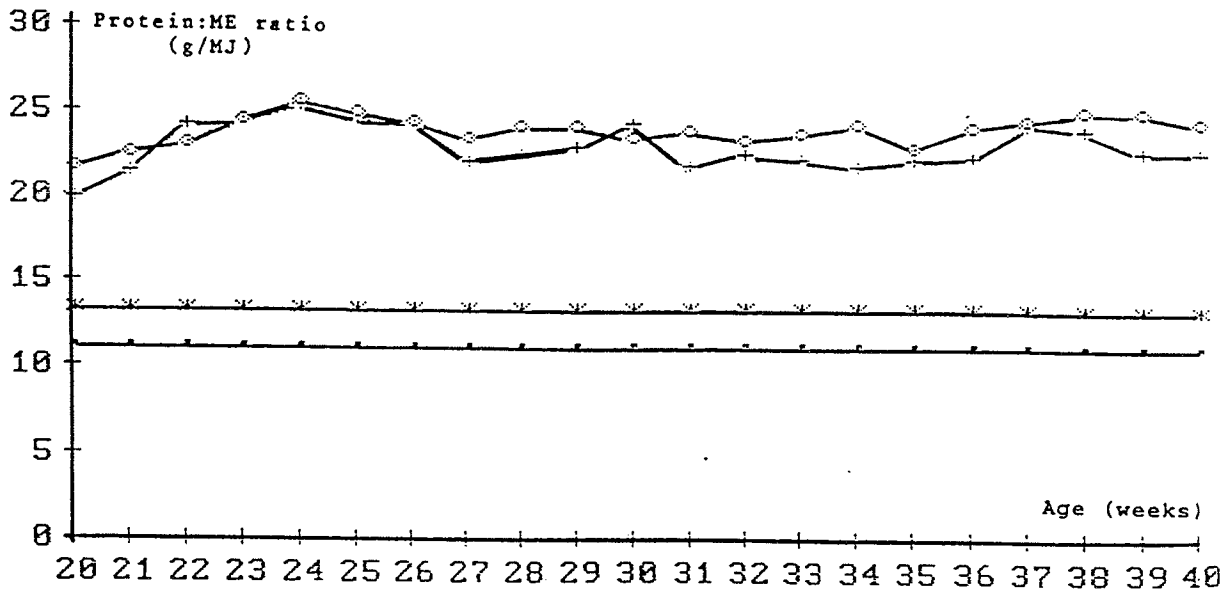


Figure 1. Response of laying hens to self-selection feeding in a 25°-35°C diurnal cycling temperature regimen (Balnave and Murtisari Abdoellah, 1990).

Long-term studies conducted by Balnave and Murtisari Abdoellah (1990) confirmed the advantages of using self-selection feeding with laying hens at high temperatures. In addition, when these workers fed a complete diet based on the ME and protein concentrations selected by hens at 25°-35°C from separate energy- and protein-rich feeds they found that hens at 25°-35°C produced an egg mass output between 19 and 40 weeks of age which was numerically superior, and non-significantly different, to that of hens housed at 10°-20°C and fed a conventionally-formulated diet. Also, for hens fed the selected diet, egg mass output at 25°-35°C was numerically less than, but not significantly different from, 10°-20°C (Table 1). The conventional diet contained 150 g crude protein and 12.0 MJ of ME/kg and the self-selection diet contained 236 g crude protein and 10.78 MJ of ME/kg. This study implies that, apart from an increased dietary protein concentration, a reduction rather than an increase in dietary energy concentration is necessary to achieve optimum egg output at high temperatures, and this perhaps explains the inconsistent responses obtained with high nutrient density diets incorporating increased energy levels. Marsden *et al.* (1987) found that a high-energy, high-protein diet was unable to maintain egg output at 30°C. Picard *et al.* (1987) reported that hens at 33°C could perform equally well on pelleted diets containing either 183 g crude protein and 11.7 MJ of ME/kg or 161 g crude protein and 9.2 MJ of ME/kg. The lower energy diet increased food consumption which gave similar intakes of energy but a 17 percent increase in protein consumption between 22 and 38 weeks of age. Therefore, the feeding of

low-energy diets at high temperatures is not necessarily a liability since it can allow increased consumption of nutrients other than energy.

Table 1. Influence of feeding a concentrated complete diet from 18 to 40 weeks of age at cool (10°-20°C) and hot (25°-35°C) temperatures (Balnave and Murtisari Abdoellah, 1990).

Variable	Diet	Cool	Hot
Food intake (g/b/d)	Conventional	95.1 ^b	74.6 ^a
	Concentrated	107.7 ^c	94.1 ^b
Protein intake (g/b/d)	Conventional	15.1 ^b	11.9 ^a
	Concentrated	27.6 ^d	24.1 ^c
ME intake (MJ/b/d)	Conventional	1.14 ^c	0.90 ^a
	Concentrated	1.16 ^c	1.01 ^b
Egg mass (g/b/d)	Conventional	38.3 ^b	31.8 ^a
	Concentrated	43.2 ^c	40.8 ^{bc}

^{a-d}Means within a measurement with no common superscripts differ significantly (P<0.05).

Balnave and Muheereza (1998) have used a repetitive intermittent lighting regimen of 3h light:1h dark (3L:1D) to improve the food intake and egg mass output of laying hens at a constant high temperature of 32°C. This lighting regimen was compared with a conventional lighting regimen of 16L:8D from 20 to 62 weeks of age. Significant increases in food intake, liveweight gain and egg weight were observed with the 3L:1D regimen and, in addition, significant age x light interactions were observed for food intake, egg production and egg mass. Hens in the 3L:1D regimen ate significantly more food to 46 weeks of age which was reflected in a numerically greater rate of lay and significantly greater egg mass output to this age. After 46 weeks of age food intake, egg production and egg mass output were similar in both lighting regimens. These results suggest that the use of intermittent lighting may be more beneficial at high, compared to thermoneutral or low, temperatures since the use of intermittent lighting regimens in non-heat stress environments has been shown consistently to decrease food intake and egg production although improving egg weight. The use of intermittent lighting regimens is a relatively new concept and contrasts with lighting standards based on older studies which are reflected in some Codes of Practice for the welfare of poultry. For example, the Australian Code of Practice (Standing Committee on Agriculture and Resource Management, 1995) recommends that "Where poultry do not have access to daylight they should be given lighting over a period of at least 8 hours per day." The 3L:1D lighting regimen provides a total of 18 h light daily.

Another way of increasing nutrient intakes, and thereby improving production at high temperatures, is to rear the birds in a cool environment prior to exposure to hot conditions during lay. Kyarisiima and Balnave (1996) found that rearing pullets at 10°-20°C before exposing them to 25°-35°C during lay significantly improved food intake and performance characteristics (Table 2).

Similar response trends were reported by Njoya (1995) in natural temperate and hot-dry climates in Cameroon. However, using a similar approach, Njoya and Picard (1994) found that pullets reared at 32°C were not acclimatised any better to this temperature during lay than pullets reared at 20°C. Pullets reared at 20°C and 60% RH and moved to either a hot-dry (32°C, 40% RH) or hot-humid (32°C, 90% RH) environment gained more body weight but showed little other beneficial effects compared with pullets maintained throughout life in these hot environments.

Table 2. Effect of a cool rearing environment on subsequent laying performance at high temperatures (Kyarisiima and Balnave, 1996).

Temperature	Rearing period	10°-20°C	25°-35°C	Significance
	Laying period	25°-35°C	25°-35°C	
Food intake (g/d)		116	106	***
Egg production (egg/hen d)		0.81	0.75	**
Egg weight (g)		54.4	52.4	**
Egg mass (g/d)		44.0	40.5	**
Food conversion (g food/g egg)		2.63	2.63	NS
Mortality (/100 hens)		4.2	12.5	*

*P<0.05; **P<0.01; ***P<0.001; NS - not significant.

There is now increasing evidence that feeding high energy diets does not necessarily counteract the effects of high temperatures on laying performance (Njoya, 1995), does not necessarily improve performance to the levels obtained with low energy-diets (Scott and Balnave, 1988b) and can result in increased mortality (El Jack and Blum, 1978). The interactions between food, protein and ME intakes, and the associated responses in egg mass output, in the study reported by Balnave and Murtisari Abdoellah (1990) show that hens fed the concentrated diet at the hot temperatures performed as well as hens fed the conventional diet at the cool temperatures although they consumed significantly less energy (Table 1). Egg mass output was related more to protein than to energy intake.

III. FATS AND OILS

Fats and oils are often considered ideal energy sources at high temperatures because of their low heat increment and the fact that they can be used to increase energy intake without elevating heat production. Njoku and Nwazota (1989) showed that the dietary inclusion of palm oil on an isoenergetic and isonitrogenous basis reduced the effect of heat stress and increased food intake, egg production, egg weight and feed efficiency.

The fatty acid composition of the dietary fat has an important influence on ameliorating the adverse effect of high temperatures on egg weight. In particular, the benefit of supplementing a layer diet with a source of linoleic acid is clearly evident from a study in which diets based on wheat or rice pollard, and containing 7 and 24 g/kg of linoleic acid respectively, were alternated every four weeks to laying hens maintained in a diurnally cycling 25°-35°C temperature environment (Balnave, 1987). The results of this study are shown in Table 3 and indicate that whereas egg weight declined each time the wheat-based diet was fed, egg weight was improved when the wheat diet was replaced with the rice pollard diet. Also, hens receiving the wheat diet continuously showed an immediate reduction in egg weight whereas those fed the rice pollard diet continuously maintained egg weight at the pre-experimental level.

Table 3. Influence of dietary linoleic acid on egg weight (g) from hens maintained at 25°-35°C over three 4-week periods (Balnave, 1987).

Dietary regimen			Pre-experiment	Week 4	Week 8	Week 12
Weeks						
1-4	5-8	9-12				
W	W	W	59.0	56.6 ^b	56.4 ^b	57.3 ^b
RP	RP	RP	58.4	57.6 ^a	58.0 ^a	58.4 ^a
W	RP	W	58.4	56.4 ^b	58.0 ^a	57.4 ^b
RP	W	RP	58.5	58.0 ^a	55.7 ^b	57.8 ^{ab}

^{a-b} Means within each week with no common superscripts differ significantly ($P < 0.05$).

IV. MINERALS

The dietary concentration of minerals is normally increased in proportion to the expected reduction in food intake at high temperatures but few studies have examined actual requirements. Most attention has been given to calcium and phosphorus which are known to influence egg shell quality.

Nordstrom (1973) reported that the reduced egg shell quality observed at high temperatures was not due to a reduced residence time in the shell gland. The use of oyster shell rather than ground limestone as the dietary calcium source has sometimes been shown to improve egg shell quality at high temperatures (Sauveur and Picard, 1987). Although blood ionised calcium concentrations are reduced during heat stress (Odom *et al.*, 1986) it is generally assumed that hyperventilation, and the associated respiratory alkalosis, is mainly responsible for inferior shell quality. The use of dietary supplements of sodium bicarbonate has generally given inconsistent results (Hughes, 1988), although ensuring that the hen consumes the bicarbonate source during the period of egg shell formation has been shown to significantly improve egg shell breaking strength (Balnave and Muheereza, 1997). In practice, this means employing repetitive intermittent lighting regimens. Advantages in egg shell breaking strength have been observed from the feeding of 10 g sodium bicarbonate/kg to hens housed in repetitive 3L:1D lighting schedules at 32°C over a 40 week laying period (Balnave and Muheereza, unpublished results) (Figure 2). Over the complete experiment the sodium bicarbonate supplement improved the mean shell breaking strength of eggs from hens in the 3L:1D regimen from 36.8 to 39.4 Newtons whereas a much smaller improvement from 32.2 to 33.2 Newtons was observed in eggs from hens in a conventional 16L:8D lighting regimen. This 7 percent improvement in the 3L:1D regimen occurred in addition to the 14 percent improvement (36.8 vs 32.2 Newtons) resulting from the use of the 3L:1D regimen.

Charles *et al.* (1978) observed that the available phosphorus requirements of laying hens was greatly increased during periods of heat stress. Usayran and Balnave (1995) found that although plasma phosphorus levels increased with increases in dietary phosphorus concentrations at both 18° and 30°C those of hens at 30°C were significantly lower than those of hens at 18°C. However, increases in dietary phosphorus adversely affected egg shell quality at both temperatures. The total phosphorus requirement on the wheat-based diets fed by Usayran and Balnave (1995) was determined as 3.2 g/kg, although the high intrinsic phytase content of wheat would have influenced this value. The results of these studies and that of Simons *et al.* (1992) show that a dietary phytase enzyme supplement of 200 phytase units/kg is sufficient to optimise the performance of laying hens fed proprietary diets.

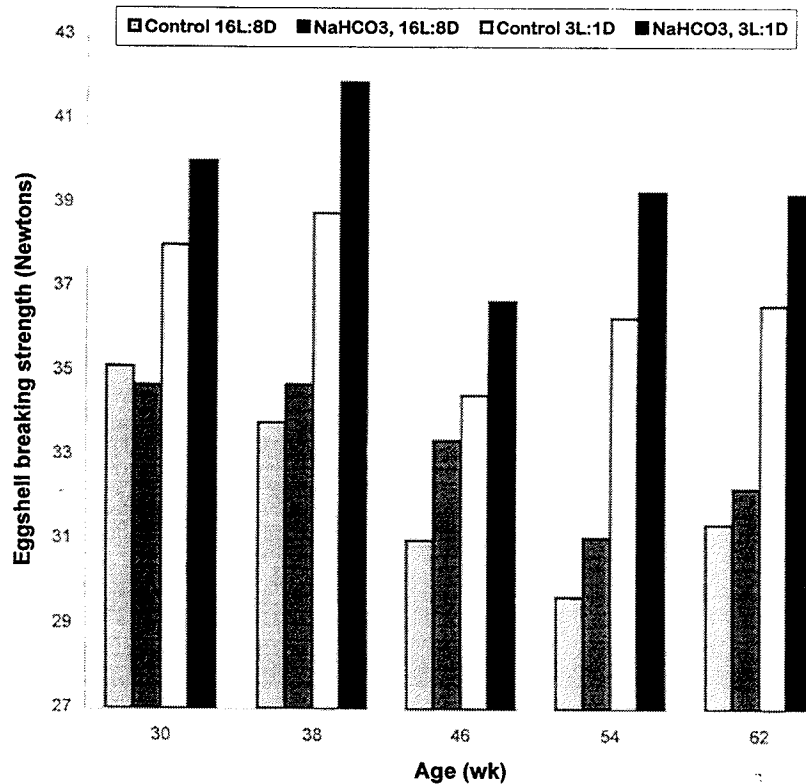


Figure 2. Effect of lighting and dietary sodium bicarbonate on shell breaking strength.

V. VITAMINS

The effect of high temperatures on the vitamin requirements of laying hens has received little attention. Most of the interest has centred on the use of ascorbic acid (vitamin C). It appears that ascorbic acid exerts its greatest influence when poultry are exposed to either an environmental or a nutritional stress (Pardue and Thaxton, 1986). Although experimental results have not always been consistent, recent studies have shown that dietary ascorbic acid supplementation levels of 100-400 mg/kg help to counteract the adverse effects of heat stress on laying hens (Njoku and Nwazota, 1989; Cheng *et al.*, 1990). Mortality is reduced and food intake, food utilization, egg production, egg weight and shell quality are improved. Recently, Khalafalla and Bessei (1997) have reported that the value of ascorbic acid supplements in the drinking water in preventing the decline in egg shell quality observed with saline drinking water (Balnave *et al.*, 1991) is only observed at high temperatures.

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FEEDING OF BROILERS AND LAYERS IN THE TROPICS OF ASIA

A.P. SINURAT

Summary

High temperatures and humidities characterize the climate in the tropics of Asia. Although this climate does not support maximum performance of broilers and layers, chicken meat and egg production are increasing in these areas. Since local feed production cannot meet requirements, imports of feed ingredients such as maize and soybean meal are also increasing.

In practice, feed formulations used in these areas follow the recommendations of the National Research Council (NRC), USA or the Agricultural Research Council (ARC), UK, although many studies have shown that the nutrient requirements of broilers and layers at high temperatures differ from those determined in cool climates. Feed supplements, such as electrolytes and vitamins, are commonly used by producers in order to alleviate the effects of high temperatures.

I. INTRODUCTION

Parts of Asia lie in the tropics, i.e. the region between 23°27' north and 23°27' south latitudes. High temperatures and humidities throughout the year characterize the climate in this area. For example, the average annual temperature in Kuala Lumpur (Malaysia) is 27.1°C with a relative humidity (RH) of 70-80% and in Jakarta (Indonesia) the equivalent data are 26.9°C and 75-85% (Yamada, 1986). Daily temperatures in Indonesia range from 23 to 35°C at low altitudes and from 20 to 30°C at high altitudes (Creswell and Hardjosworo, 1979). Similar high temperatures and humidities also occur in other tropical countries as indicated by Azahan (1996) in Malaysia, Devegowda (1995) in India and Muangcharoen *et al.* (1992) in Thailand.

Chickens reared in these areas are exposed to a climate above the comfort zone every day. A study conducted by Sinurat and Habibie (1991) showed that for about 8 to 12 hours each day the temperature in the chicken house was above the recognised thermoneutral zone (27°C). This climate is without doubt the cause of the performance of poultry in tropical countries being inferior to that in temperate countries, as illustrated by the performance data in Table 1.

Poultry is an important source of protein for people in this region. The poultry population in 1995 was 1558 million birds which produced 3664000 tons of chicken meat and 3494518 tons of eggs (Table 2). Although native chickens and ducks are popular, the exotic birds (broilers and layers) contribute in a major way to the protein consumption of society. The poultry population is growing rapidly in this region. According to FAO (1996) the chicken population grew between 6% and 14 % from 1994 to 1995. Although the consumption of chicken meat and eggs are increasing, their per capita consumption is low compared to countries like Australia.

Research Institute for Animal Production, PO Box 221, Bogor 16002, Indonesia.

Nevertheless, some of the countries in this region are already exporting chicken meat. In 1994 Indonesia, Malaysia and Thailand exported 1103, 6578 and 158941 tons of chicken meat, respectively (FAO, 1995a).

Table 1. Comparison of the performance of broilers and layers in the tropics and temperate countries.

Parameters	Tropics	Temperate
<i>Broilers</i> ^a		
Country	Thailand	Netherlands
age at harvest (d)	42-44	43
Live weight at harvest (kg)	1.8 - 2.0	1.85
Feed conversion ratio	1.9 - 2.1	1.86
Mortality (%)	5.70	4.60
<i>Layers</i>		
Country	Indonesia ^b	Canada ^c
Strain	Dekalb	Dekalb
Egg prod. 34 weeks (% HD)	74.6 - 85.0	89.6
Feed intake (g/d)	100.3 - 113.4	97.7
FCR	2.47 - 2.68	1.88
Egg weight (g)	58.6 - 65.8	58.0

Sources: ^a Anonymous (1996); ^b Sinurat *et al.* (1996) ^c Leeson *et al.* (1997).

Table 2. Poultry population and production of some countries in Asia in 1995.

Country	Chicken population (million)	Poultry meat production (x 1000 ton)	Egg production (ton)
Bangladesh	1	106	82000
India	610	578	1540000
Indonesia	650	759	452636
Malaysia	100	700	354869
Myanmar	9	29	37000
Philippines	3	403	305000
Sri Lanka	10	54	48950
Thailand	80	857	538063
Vietnam	95	178	136000
Total	1558	3664	3494518

Source: FAO (1996).

II. FEED INGREDIENTS AND PRODUCTION

The increasing poultry population has increased the demand for feed in this region. In 1995 as much as 18.3 million tons of poultry feed was manufactured in this region (Best and Gill, 1996) with India producing the largest quantity (6.3 million tons) followed by Indonesia (3.5 million tons) and Thailand (3.5 million tons). Other countries each produce less than 2 million tons of feed per year (Table 3). The diets for broilers and layers are mainly composed of maize, rice bran, wheat bran/pollard, soybean meal, fish meal, coconut meal, palm kernel

meal, coconut oil, crude palm oil, meat and bone meal, minerals and vitamins. Cassava meal, although produced locally in large quantities, is not commonly used in commercial ration formulation due to its low concentration of nutrients other than energy. Protein enrichment by fermentation processes, as reported by Kompang *et al.* (1995), may increase the inclusion of cassava in commercial diets in the future. There are many other feed ingredients used by small farmers that are available locally such as kapok seed meal, leaf meal, sago meal, etc. These ingredients are not commonly used by commercial feed millers. These ingredients, and their nutritional values, have been reviewed by Ravindran and Blair (1991, 1992, 1993).

Some feed ingredients are produced locally such as maize, fish meal, rice bran, palm kernel meal, crude palm oil and coconut oil. The amount of ingredients produced, however, cannot meet the increasing need for manufactured feed and, therefore, reliance has to be placed on imports (Table 3). Maize and soybean meal are the major components of commercial poultry feed in this region. Therefore, the volume of imports of these two ingredients are increasing. The volume of imports will probably continue to increase in the future, except for maize, since Indonesia and Malaysia, the largest importing countries, have started to increase local maize production in the last two years.

Table 3. Poultry feed production and imports of some ingredients by countries in Asia.

Country	Poultry feed ^a production (million ton)	Volume of Imports in 1994 (ton) ^b			
		Maize	Soybean meal	Fish meal	Meat and bone meal
India	6.3	-	200	6954	-
Indonesia	3.5	1118300	498590	247918	189375
Thailand	3.5	34900	902710	206375	26841
Malaysia	1.8	1968800	465070	25935	16482
Philippines	1.8	900	65507	112895	13973
Vietnam	1.5	400	-	-	-
Bangladesh	0.2	148	-	-	-
Myanmar	0.1	-	-	-	-

Sources: ^a Best and Gill (1996); ^b FAO (1995a).

III. FEED FORMULATION

Almost all the strains of commercial broilers and layers that exist in tropical countries were originally bred in temperate countries and best perform in a 'cool' climate. Hence, it is not surprising that feeds for broilers and layers are formulated to meet the nutrient requirements as determined in temperate climates. The most common guidelines that are used in practical diet formulations are the NRC and the ARC recommendations. As an example, the dietary nutrient specifications for broilers and layers produced by a commercial feed miller in Indonesia is shown in Table 4 (Hutagalung, personal communication). The nutrient specifications for broilers are very close to the recommendations of the NRC (1984), except that a lower ME is applied. The layer diet, however, tends to have a higher protein than the levels recommended by the NRC (1984). Similar specifications have also been applied in commercial poultry feed in India (Saxena, 1992).

Many researchers have shown that the nutrient requirements of broilers and layers in hot climates are different to those in cool or moderate climates. Balnave (1996) summarized results of studies on the nutrition of broiler chickens and laying hens at high temperatures. The results suggested that broilers kept at high temperatures should be fed higher energy diets

while simultaneously reducing dietary amino acid concentrations. Other options included increasing the dietary arginine:lysine ratio, selecting specific forms of dietary methionine and dietary methionine:total sulphur amino acid ratios and supplementing the diet or drinking water with sodium bicarbonate. Also, laying hens should be fed diets with a higher protein:energy ratio compared with those recommended for moderate/cool temperatures.

Table 4. Nutrient specifications of feed for broilers and layers produced by a commercial feed miller in Indonesia.

	Protein (g/kg)	ME (MJ/kg)	Prot:ME ratio (mg/KJ)	Lysine (g/kg)	Methionine (g/kg)
<i>Broiler diet:</i>					
Starter	200-230 (230)	12.55-12.97 (13.39)	160-17.7 (17.2)	12.5 (12.0)	5.0 (5.0)
Finisher	180-220 (200)	12.55-13.39 (13.39)	14.3-17.0 (15.1)	11.5 (10.0)	4.5 (3.8)
<i>Layer diet:</i>					
Starter (1-6 w)	180-200 (180)	11.92-12.55 (12.13)	15.1-16.0 (14.8)	9.8 (8.5)	4.0 (3.8)
Grower (6-14 w)	170-190 (150)	11.51-12.13 (12.13)	14.8-15.8 (12.4)	8.5 (6.0)	3.0 (2.5)
Developer (14-20 w)	140-160 (120)	10.88-11.51 (12.13)	12.7-12.9 (9.8)	7.0 (4.5)	3.0 (2.0)
Layer production	160-180 (150)	11.09-12.34 (12.13)	14.3-15.8 (11.9)	8.0 (6.4)	3.0 (3.2)

Values in brackets are recommendations of NRC (1984).

Reductions in feed intake and, hence, the intake of all nutrients by birds kept at high temperatures is believed to be the primary cause of the inferior performance of chickens in the tropics. Therefore, attempts to improve the performance of poultry in tropical countries have concentrated on increasing the intake of nutrients or by increasing the nutrient density of the feed. In Malaysia, Hamid and Jalaludin (1986) showed that increasing the dietary energy from a low (10.1 MJ ME/kg) to a medium (11.3 MJ ME/kg), but not to a high (12.6 MJ ME/kg) concentration, improved hen day egg production, egg weight and feed conversion. All diets were formulated to be similar in protein content (180 g/kg) and the temperature during the experiment was 24-26°C (minimum) and 34-36°C (maximum). In India, low energy layer diets (10.5 MJ ME/kg) with 170 g/kg crude protein are commonly used (Devegowda, 1995). Farrell (1995) and Balnave (1996) have indicated the benefits of self selection or free choice feeding in improving the performance of layers at high temperatures. Both authors also recommended higher dietary protein:energy ratios for layers in hot temperatures as compared to conventional rations.

Raghavan (1993) reported that broilers consuming a high density diet from day old through to the finishing period grew faster with a better feed conversion than those on low density starter and finisher diets. However, the nutrient levels of the high density diet were similar to the starter diet specifications recommended by NRC (1984), as shown in Table 5.

Table 5. Performance of broilers fed diets with different nutrient densities under tropical conditions.

	Low density		High density	NRC (1984)	
	Starter	Finisher	(Starter & Finisher)	Starter	Finisher
<i>Diets specification:</i>					
Crude protein (g/kg)	221	190	241	230	200
ME (MJ/kg)	12.34	12.55	13.39	13.39	13.39
Lysine (g/kg)	10.8	9.5	12.0	12.0	10.0
Meth. + Cys. (g/kg)	8.3	7.4	9.2	9.3	7.2
<i>Performance to 7 wks:</i>					
Feed intake (kg)	4.275		4.315	3.840	
Live weight (kg)	1.763		2.008	1.923	
FCR (g:g)	2.42		2.178	2.00	

Source: Raghavan (1993).

IV. FEED SUPPLEMENTS

Although most commercial broilers and layers are fed complete diets, feed supplements are also commonly given by producers. The feed supplements are given in order to improve profitability through an improvement in body weight gain, egg production, feed efficiency or by reducing mortality. The kind of feed supplements used varies from one producer to another but, in general, they can be classified as antistress, growth promotant, enzyme or probiotic compounds.

In relation to the high temperatures in tropical countries, antistress agents are widely used for broilers and layers through feed or drinking water supplementation. The feed supplement mainly consists of minerals/electrolytes and vitamins. Sinurat *et al.* (1992) reported that a high dietary electrolyte balance (250-450 meq/kg *vs.* 125 meq/kg) significantly reduced the body temperature of broilers kept at high temperatures (26-33°C). Further study (Sinurat, unpublished data) has shown that hens reared at a temperature of 20-32°C and a RH of 70-78% and given a commercial electrolyte supplement in the drinking water improved egg production and feed conversion (Table 6).

Table 6. The effect of a commercial electrolyte supplement on the performance of laying hens in the tropics.

Parameters	Control	Supplement
Feed intake (g/d)	110.60	114.70
Egg production (% HD)	78.90	82.90
Egg weight (g)	58.04	60.55
FCR (g:g)	2.449	2.293
Shell thickness (mm)	35.13	36.33

Although vitamin C (ascorbic acid) is naturally synthesized by chickens, its usage as an antistress agent for poultry in the tropics is not unusual. Harimurti (1992) showed that vitamin C can alleviate the effects of heat stress on laying hens as shown by a reduction in leucocyte numbers in the blood and an improvement in performance (Table 7). Feeding of guava fruit, a natural source of vitamin C, however, did not significantly improve the performance of the hens although it did significantly reduce leucocyte numbers.

Table 7. The effect of ascorbic acid (AA) supplementation on the performance and blood leucocyte numbers of laying hens kept in the tropics.

Parameters	Control (C)	C + 48 mg AA/day	C + 72 mg AA/day	C + guava fruit
Feed intake (g/d)	117.3	119.8	121.8	118.7
Egg production (% HD)	67.6 ^a	79.5 ^b	79.2 ^b	74.1 ^{ab}
Egg weight (g)	59.3	60.6	61.1	60.2
Feed conversion (g:g)	2.97 ^a	2.51 ^b	2.53 ^b	2.68 ^a
Leucocytes (x10 ⁶ /mL)	17.1 ^a	9.9 ^b	9.3 ^b	10.2 ^b

The guava fruits provided approximately 72 mg vit C/d.

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SOME ASPECTS OF AGEING FOR EGG PRODUCTION OF LEGHORNS

R.W. FAIRFULL^{1,2}, L.-E. LILJEDAHN³, R.S. GOWE⁴, I. McMILLAN⁴
and L.B. ASSELSTINE¹Summary

White Leghorn strains were crossed reciprocally in a complete factorial mating producing 6 pure strains and 30 strain crosses which were kept for two laying cycles: 133-496 days of age (d) and 547-909 d (364 and 363 d, respectively). Hens were individually housed for lay in four two-tiered batteries of cages. Strain additive effects (A_i), strain sex-linked effects (Z_i), strain-cross heterotic effects (h_{ij}) and residual effects were calculated using regression. Phenotypic performance for egg production decreased with age. The magnitude of genetic effects increased with age as did their variation. Environmental variation also increased with age. Heterosis had a generally positive influence on late performance, which was greater than that of other genetic effects. Overall, these results suggest that genetic expression does not become fixed and that the use of performance data from late in the first laying cycle or from the second cycle would allow the utilization of genetic variation not available earlier.

I. INTRODUCTION

Many people portray the life cycle of organisms as an extremely complex set of events. In reality, life cycles are very simple when you eliminate unnecessary detail: only two events are especially important, *birth* and *death*. Everything between them comprises a process called *ageing*. For example, are sexual maturity and reproduction important? Yes, they are, after all, ageing processes. Ageing is the sum of many genetic, physiological, developmental and environmental (internal and external) processes. It is not a single isolated event and does not result from a single mechanism. Most physiological systems are affected by ageing, for instance the neuro-endocrine, the immune, and the reproductive systems. Ageing is characterized mainly by declining vigor, reduced viability and increasing environmental sensitivity.

On an individual basis is ageing important? Possibly not: there is overhead cost in producing an independent adult and on an individual basis reducing the reproductive value of each adult in a retrogressive process does not seem worthwhile. On a population basis, however, replacement units must be continuously available and the ability of a population to react to crisis or adapt would be reduced without ageing to help get rid of those obsolete models. In evolutionary terms, it allows for genetic flexibility. In a population sense, it allows for growth and change.

Theories of ageing revolve around two central themes: i) ageing results from an accumulation of random events; and ii) ageing is a programmed, time-dependent process.

¹ Centre for Food and Animal Research, Agriculture Canada, Ottawa, Ontario K1A 0C6, Canada.

² Present address: ARGO Breeding Co., Nacogdoches, Texas, 75963-1940, U.S.A.

³ Swedish University of Agricultural Sciences, Uppsalla, Sweden.

⁴ Animal and Poultry Science, University of Guelph, Guelph, Ontario, N1G 2W1, Canada.

Some theories of ageing (see Esser and Martin, 1995) are: 1) the somatic mutation theory - an accumulation of random, deleterious somatic mutations; 2) the error catastrophe theory - an accumulation of random, deleterious mutations resulting in transcription and translation errors; 3) mutation accumulation theory - an accumulation of random, deleterious mutations expressed only late in life, modifier genes delay expression; 4) the antagonistic pleiotropic theory - alleles favorable to early fitness and deleterious to late fitness become fixed; and 5) the disposable soma theory - maximum fitness balances somatic maintenance and reproduction. In evolutionary biology, a circular theory has emerged. Specifically, assuming genetic variation in life patterns, if the intensity of natural selection for age specific fitness declines with age, then senescence developed as a by-product of evolution. There are several problems with evolutionary explanations: the theory requires genes with high early fitness with late low fitness trapped by natural selection. However, natural selection has little effect late in life and there is no satisfactory mechanism for random events.

Information on ageing processes does not always aid understanding of genetic mechanisms of ageing in a straightforward manner. The rate of protein synthesis declines with age, but there are no qualitative differences with age (Arking and Dundas, 1989). The ability of the cells to communicate with each other and receive information from the environment (signal transduction) alters with age (Roth, 1990). This does not seem to be related to the concentration of receptors as some decrease with age, other remain unchanged, while still others increase. However, the ability of steroid hormone receptors to bind to DNA acceptor sites declines with age. The expression of structural genes varies with age (Thaker *et al.*, 1993): some have high early expression then decline; others have low early expression that increases to a maximum then decline; still others steadily increase expression to a high level late in life. In humans the most probable causes of ageing are genetic damage (Strehler, 1989) and genetic instability (Slagboom and Vijg, 1989). Also, there are several examples of specific genes directly causing ageing in several species, for example Werners syndrome in humans. Individual variation in regulation exists (Cinader, 1989a,b): activation of genes later in life and delayed manifestation of activated genes. In mice, inactivated X-chromosome genes are activated later in life (Cattanach, 1974; Wareham *et al.*, 1987). What conclusions can be drawn from this information? First, ageing and its effects are very complex. Second, these observations support several theories of ageing. Third, no single theory of ageing is adequate.

Heterosis is the deviation between the cross mean and the mean of the parents. Genetically, it is due to interactions: among alleles (dominance) or among different genes (epistasis). Heterosis seems to provide buffering (homeostasis). In laying hens, it is larger under adverse conditions and in traits associated with fitness (Fairfull *et al.*, 1987; Fairfull, 1990).

II. MATERIALS AND METHODS

The information reported here derives mainly from a study conducted with six SC White Leghorn strains from three genetic base populations developed at the Animal Research Centre in Ottawa. All the six strains were selected primarily for egg production from housing to 273 days of age, and for egg weight, eggshell strength, viability, fertility, hatchability and egg quality traits. The strains were crossed reciprocally in a factorial design with 56 males and 112 females used to produce each of six pure strains and 30 strain crosses.

All chicks were reared in four batteries of a 3-tier cage system in a windowless house to 132 days of age. At 133 days of age, the pullets were housed one per cage (20.3 cm wide) in four batteries of a 2-tier cage system in a windowless house. For each cross and pure

strain, 57 pullets were housed. All mash diets were fed *ad libitum* throughout the study. Birds were vaccinated for Marek's disease, avian encephalomyelitis, bronchitis and Newcastle disease. Artificial light was provided for 24 h after hatch and then reduced to 6 h daily until 132 days of age. At housing, light was augmented to 8 h daily and increased by 30 minutes per week to a maximum of 16 h daily. At the end of the first egg production cycle (496 days of age), all birds were induced to molt (Fairfull, 1982). The second egg production cycle started at 546 days of age when 16 h light daily was resumed.

Egg production was recorded 5 d per week from 133 to 496 days in the first cycle and from 547 to 909 days of age in the second cycle. For analysis, the egg record of each hen was divided into four week periods that started with the week the first egg was laid, so that each cycle had 11 periods. Mortality was recorded daily and all dead hens were necropsied. Hens that died as the result of an accident were removed from consideration. Strain additive effects (A_i), strain sex-linked effects (Z_i) and strain-cross heterotic effects (h_{ij}) were estimated by regression (Robison *et al.*, 1981) using the model:

$$Y_{ijk} = \mu_p + \frac{1}{2}A_i + \frac{1}{2}A_j + Z_i + \frac{1}{2}h_{ij} + e_{ijk}$$

where Y_{ijk} = the observation on the k^{th} individual of a mating between a strain i sire and a strain j dam; μ_p = mean of the p pure strains, $\frac{1}{2} = 0$ when $i = j$ or $\frac{1}{2} = 1$ when $i \neq j$, and e_{ijk} = a random residual effect.

III. RESULTS

(a) Performance

Egg production decreased within cycles and across cycles for both crosses and pure lines (Liljedahl *et al.*, 1984,1998). After an initial small increase from the first to the second period in the first cycle, egg production declined steadily. In the second cycle, egg production recovered somewhat, then decreased to the end of the cycle (Figure 1). In the second cycle, the rate of decline was greater than that of the first cycle.

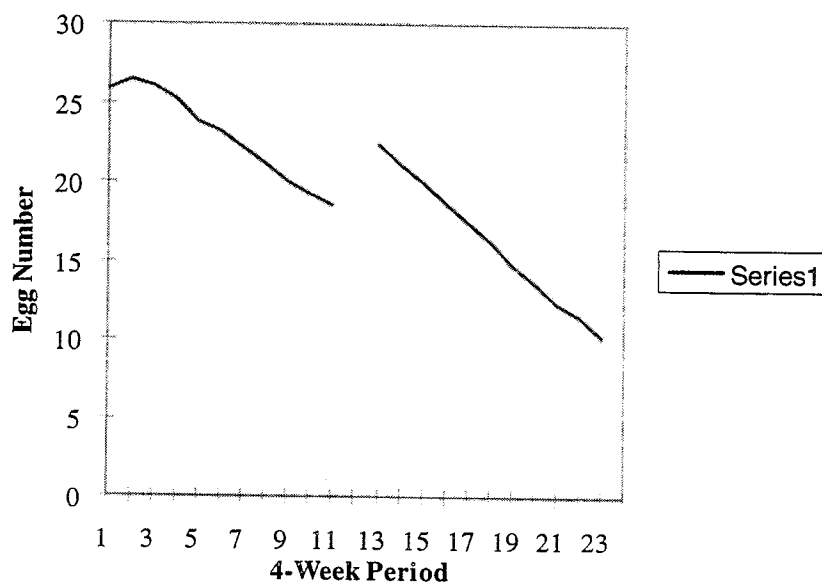


Figure 1. Mean egg production by period.

(b) Additive genetic effects

Additive genetic variation for egg production increased with age within each cycle and across cycles (Figure 2; Liljedahl *et al.*, 1984, 1998). Also, the magnitude of strain somatic (A_i) and sex-linked (Z_i) additive genetic effects for egg production increased with age within each cycle and across cycles. Patterns of age changes in total additive genetic effects (A_i+Z_i) varied widely from strain to strain (Figure 3) and differences among strains increased notably with age.

(c) Heterosis

Generally, strain cross heterotic effects (h_{ij}) increased with age in each of the two laying cycles and from cycle 1 to cycle 2 (Figure 4). Heterosis was consistently large and positive although three crosses exhibited negative heterosis in some periods. In addition, the large differences among strain crosses for heterotic effects increased clearly across ages and the variance of heterotic effects increased with age (Liljedahl *et al.*, 1998). Thus, genetic interactions seem to be more important to late as opposed to early reproduction.

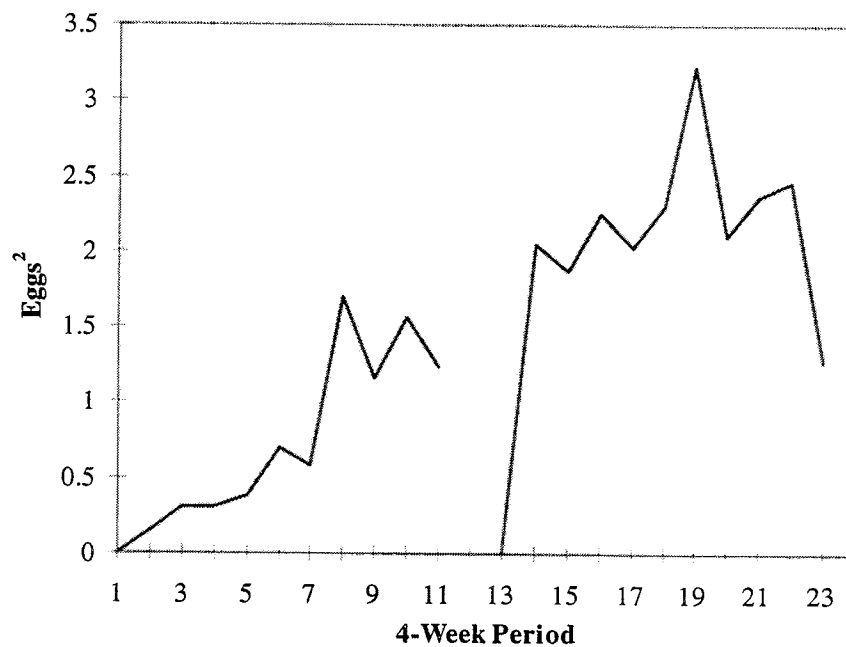


Figure 2. Additive genetic variance by period.

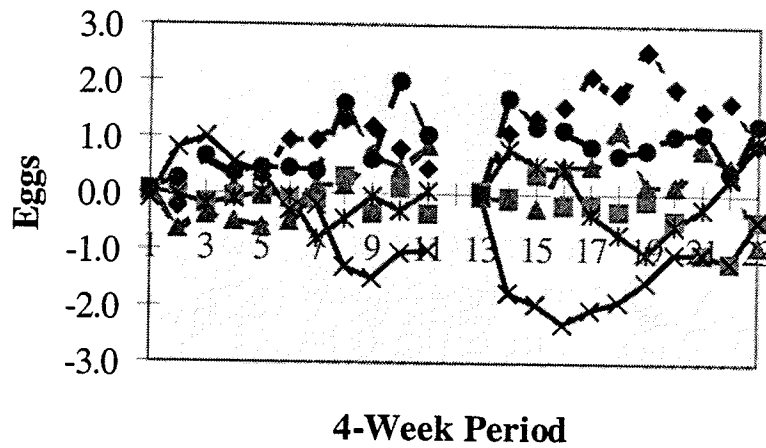


Figure 3. Strain additive (A_i+Z_i) effects by period.

(d) Environmental effects

Environmental variation for egg production increased with age within each cycle and the increase in the second cycle was greater than that of the first cycle (Figure 5; Liljedahl *et al.*, 1984, 1998). The environmental variance increased in an almost linear fashion across the first laying cycle. In the second cycle, the increases became more pronounced in the earlier periods, but rose to a maximum by about the 20th period after which it declined slightly (Figure 5).

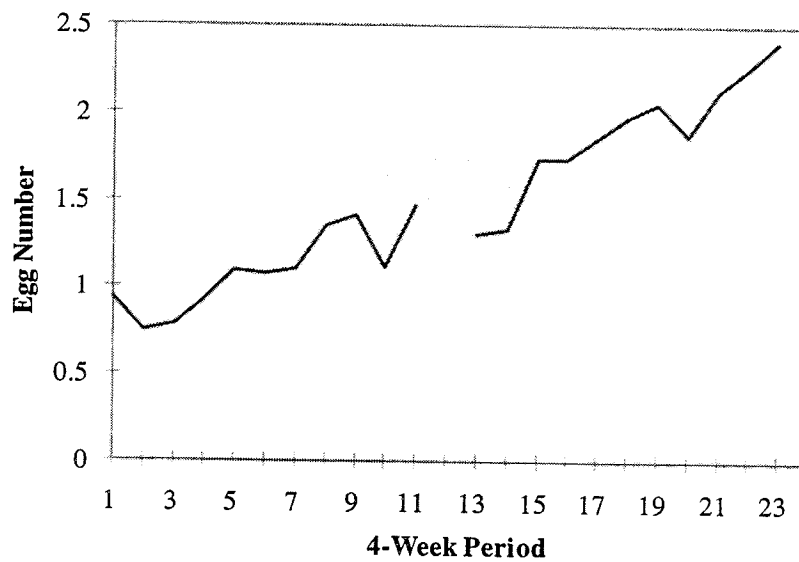


Figure 4. Heterosis for egg production by period.

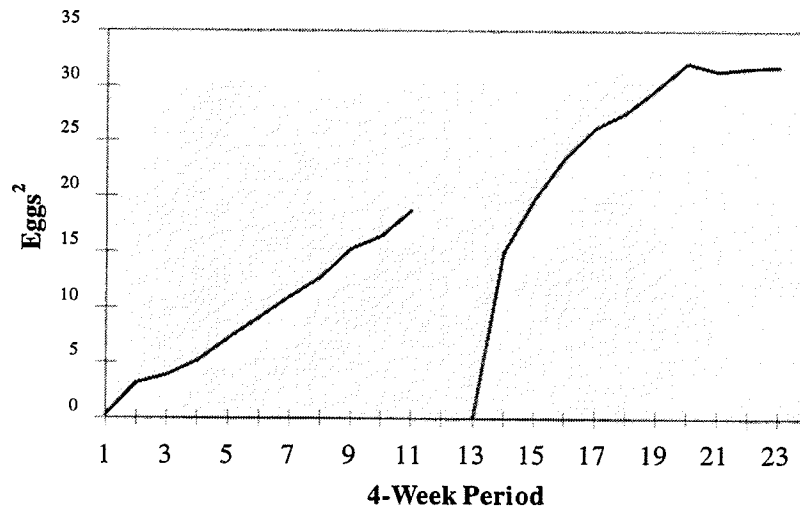


Figure 5. Environmental variance by period.

IV. DISCUSSION

(a) Strategies of ageing

The great variation among genotypes across ages in genetic effects implies that genotypes apply different genetic strategies to reduce the negative impact of ageing. Both additive and non-additive genetic effects are important in ageing; however, heterotic effects are clearly larger than other genetic effects and exert more influence over the effects of ageing. Genetic interactions increase in importance with respect to reproduction as chickens increase in age. Basal metabolic rate is lower and reproductive performance higher in crosses. Heterosis is greater in fitness traits which are influenced by both dominance and epistasis (Fairfull *et al.*, 1987). Also, heterosis is greater under adverse environmental conditions. The increasing heterosis with age may reflect in part a deteriorating internal environment. Heterosis seems to provide a capacity to buffer (reduce) the deleterious effects of ageing. This might be expected as heterozygosity increases flexibility: **2n** versus **n** pathways. Conclusion: gene interactions are an important genetic mechanism in ageing and extended performance.

These results and other recent findings (see Gowe and Fairfull, 1985,1986) cast doubt on the validity of using only early records in selection. The variation of genetic effects for egg production and other fitness traits rises markedly with age. This suggests that genetic expression does not become fixed and that consideration of reproductive traits at later ages may allow the utilization of genetic variation not available earlier.

(b) Influence of the Environment

The environment has a great influence on egg production at all ages. Also, increases in environmental variation with age are proportionately greater than those of genetic effects. In examining causes of mortality, only renal failure showed a significant age trend: it was higher at the end of each cycle. This suggests that diet may be an environmental factor that is

important in extended performance. Perhaps, tailoring layer diets to the physiological needs of hens at various stages of life could reduce negative environmental influences and improve extended performance.

V. CONCLUSIONS

First, gene interactions are very important in buffering the deleterious effects of ageing. Second, different genotypes use different strategies to diminish the effects of ageing.

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WELFARE ASPECTS OF BEAK TRIMMING IN POULTRY

M.J. GENTLE

Summary

This paper reviews current research on the welfare problems associated with beak trimming in poultry. The benefits to the bird are discussed, together with the adverse effects of potential acute and chronic pain, and sensory loss. The causal mechanisms for feather pecking are not understood and at present there is no clear hypothesis to explain the effectiveness of beak trimming in reducing feather pecking and cannibalism. A case can be made for the continuation of beak trimming on welfare as well as production grounds provided it is performed only once in young chicks. In the long term trimming should be phased out and undesirable behaviour controlled by environmental means and genetic selection of commercial stock which do not engage in damaging pecking.

I. INTRODUCTION

Beak trimming, variously called debeaking or partial beak amputation, is a procedure widely used by the poultry industry for reducing the incidence and harmful effects of feather pecking, aggressive pecking and cannibalism. It has been a contentious issue ever since the publication in the UK of the report of the Brambell Committee in 1965, in which it was criticized on two main grounds: that the procedure itself resulted in pain and that it deprived the birds of full use of its 'most versatile member' (Brambell, 1965). Thirty years has now passed since the report was published, and it is generally accepted that we still do not have laying stocks available which can be confidently housed in large flocks on the floor without exposing them to the risk of a major and damaging outbreak of cannibalism. In considering the welfare aspects of beak trimming we need to consider the costs and benefits to the animal rather than the producer, although the prevention of cannibalism benefits both. There are however occasions where difficult decisions have to be made. For example feather pecking can be more easily controlled in cages than in more extensive systems, yet it is argued by many of the animal welfare organizations (UK, Farm Animal Welfare Council, Royal Society for the Prevention of Cruelty to Animals) that the welfare of the birds is better served in extensive systems even though birds have to be beak trimmed. This review presents some of the recent work on the behavioural and physiological consequences of beak trimming.

II. BENEFICIAL EFFECTS FOR THE BIRD

It is clear that reduced mortality rates in beak trimmed birds (Denbow *et al.*, 1984; Lee and Craig, 1991; Struwe *et al.*, 1992; Grigor *et al.*, 1995), reflecting decreased cannibalism, implies less suffering for individual birds. Feather pecking is also reduced and beak trimmed flocks have better plumage (Hughes and Michie, 1982; Grigor *et al.*, 1995). Considerable force is required to remove feathers with values of 400 to 750 g reported for dorsal feathers in White Leghorn hens (Ostmann *et al.*, 1963) and 2 to 4 kg to remove the primary remiges (Lucas and Stettenheim, 1972).

Roslin Institute, Roslin, Midlothian, EH25 9PS U.K.

These forces are considerably in excess of the 2 to 5g required to activate the mechanothermal and high threshold mechanical receptors (nociceptors) which signal noxious stimulation of the skin (Gentle, 1989). In a study where a variety of physiological (ECG, EEG, BP) and behavioural parameters were examined following feather removal there was clear evidence that it is painful (Gentle and Hunter, 1990) and therefore feather pecking constitutes a welfare problem. In addition to reduced feather pecking there are reports of fewer aggressive interactions when group-housed birds are beak trimmed (Eskeland, 1977); the reduced social disturbance is especially obvious at high stocking densities (Eskeland, 1981).

There is evidence that under certain environmental conditions, beak-trimmed birds may be under less stress; e.g. in some studies trimmed laying birds were less fearful than untrimmed ones, both in multi-bird cages and in floor pens (Lee and Craig, 1991), but in other studies there was no effect of beak trimming on fearfulness (Kuo *et al.*, 1991). Furthermore, cage-reared (but not litter-reared) beak-trimmed birds had smaller adrenal glands than controls (Struwe *et al.*, 1992), suggesting less stress.

III. ADVERSE EFFECTS ON THE BIRD

These can be divided into three categories of effects: acute pain, chronic pain and sensory deprivation.

(a) Acute pain

There is no comprehensive definition which would enable the unambiguous determination of whether or not an animal is in pain and there is no reliable universal indicator of pain. What can be done, however, is to compare a range of physiological and behavioural measures with those changes which are associated with pain in humans, and thereby arrive at an estimate of the probability of pain in a given situation. Zimmermann (1986) has proposed a working definition of pain in animals:

"Pain in animals is an aversive sensory experience caused by actual or potential injury that elicits protective motor and vegetative reactions, results in learned avoidance, and may modify species specific behaviour, including social behaviour".

Acute pain lasts for seconds to days and follows nociceptive stimulation or injury and when healing is complete pain is no longer present. A number of studies have shown that the beak of the chicken is well supplied with receptors which are preferentially sensitive to tissue damage, i.e. nociceptors (Roumy and Leitner, 1973; Breward, 1985; Gentle, 1989). In addition to their presence in the beak, nociceptors have also been reported in the buccal cavity (Gentle and Hill, 1987; Gentle, 1979). It would therefore follow that the tissue damage resulting from beak trimming is likely to be acutely painful. Electrophysiological recordings from the sensory nerves in the beak during and immediately after amputation using a heated blade (Gentle, 1991) has shown that the sensory fibres respond just before the blade makes contact with the beak. During amputation and for a period of from 2 to 48 seconds after amputation, there is a massive injury discharge in the sensory fibres which is likely to be responsible for the acute pain produced at the time of amputation.

A recent study of beak trimming in turkeys (Grigor *et al.*, 1995) has suggested that this acute pain may depend on the age at which the birds are trimmed and the method of beak trimming. In this study 3 different methods of trimming were used; cutting with secateurs, a heated blade debeaker or a high voltage electrical discharge (Bio-beaker, Sterwin Laboratories, Millsboro, Delaware, USA). No behavioural evidence of acute pain during trimming using secateurs was observed in turkeys trimmed at 6 or 21 days of age. Trimming with the heated blade produced escape behaviour and vocalisation suggesting they perceived the procedure as painful. Vocalisation and escape behaviour were also observed with the Bio-beaker when used

on 1-d-old chicks. Although there were some slight behavioural differences between the control and the beak trimmed birds there was no clear evidence of pain-related behaviours such as beak guarding in the trimmed birds at any period after trimming. A similar result was found when beak trimming laying hens with either secateurs or hot blade debeaker at 1- or 10-d-old (Gentle *et al.*, 1997).

In these two recent studies (Gigor *et al.*, 1995; Gentle *et al.*, 1997) anatomical studies were also performed on the beaks up to 42 d after trimming (Gentle *et al.*, 1995; Gentle *et al.*, 1997). All methods resulted in the loss of a significant amount of beak tissue. By 42 days after trimming the beak had healed with extensive regrowth, including bone and cartilage formation, and the pattern of regrowth was similar after all trimming methods. In the normal bird the dermis at the tip of the upper and lower beaks contains large numbers of nerve fibres and sensory receptors, but in the beak-trimmed birds the dermal tissue, although well supplied with blood vessels, was devoid of afferent nerve fibres and sensory endings. The neural regrowth following trauma matched that of the surrounding tissue such that there was no extensive scar tissue or neuroma formation. A similar absence of neuromas in the beaks of adult hens was reported after trimming of young birds (Lunam *et al.*, 1996; Dubbeldam *et al.*, 1995) but neuromas were present in the beaks of adult hens which had been severely trimmed at hatch (Lunam *et al.*, 1996). The neural and sensory innervation of the beaks of adult birds trimmed as young chicks showed that there was a clear loss of sensory corpuscles in the trimmed beak compared to the normal beak and there was a high density of small myelinated and unmyelinated fibres, i.e. probably predominantly representing A delta and C fibres (Dubbeldam *et al.* 1995). This change in the sensory innervation of the beak is reflected in a reduction of large nerve fibres in the trigeminal nerve, reductions in the number of large ganglion cells in the trigeminal ganglion, and a reduction in the volume of the principal sensory trigeminal nucleus in the brain (Dubbeldam *et al.*, 1995).

Beak trimming in adult hens, unlike chicks, produces evidence of pain related behaviours. These behaviours can be grouped as beak guarding behaviours and consist of a reluctance to use the beak for non-essential activities such as preening or environmental pecking. In one experiment beak guarding behaviour was measured by counting the number of pecks the birds delivered to an attractive visual stimulus before and again 6, 26 and 32 hours after partial beak amputation (Gentle *et al.*, 1991). At 6 hours after amputation the birds continued to peck the same number of times at the stimulus but by 26 hours after amputation there was a significant reduction in pecking indicating pain. These results point to the presence of a pain-free period immediately following amputation, which may last in some birds for as long as 26 hours, after which the birds appear to be in considerable pain. Some recent unpublished observations from our laboratory have confirmed this pain-free period. Birds were trained to peck at an operant key for a food reward. Immediately after amputation the birds continued to peck at the key for food but on the following day, the birds made only tentative pecking movements at the key and were unwilling to peck hard enough to operate the mechanism. Similar pain-free periods are seen in humans after major traumatic injuries especially after full-thickness burns (Robertson *et al.*, 1985; Stein and Stein, 1983). An explanation for this pain-free period may lie in the responses of sensory afferents. Although the heated blade produced a massive injury discharge in the sensory fibres of the beak for several hours after this discharge there was no further abnormal neural activity in the sensory fibres running to the amputated stump. This absence of abnormal activity in the sensory afferents could provide a mechanism to explain this pain-free period (Gentle, 1991).

(b) Chronic pain

Beak trimming in adult birds presents a very different picture to beak trimming in chicks and there is evidence of possible chronic pain in the adult animal. Behavioural changes which could be interpreted as indicative of pain have been observed for long periods after trimming. For at least 5 weeks after amputation there is a significant reduction in the use of the beak for non-essential activities such as preening and exploratory pecking (Duncan *et al.*, 1989). Other non-beak-related activities were also affected with the birds showing persistent increases in the time spent inactive (Duncan *et al.*, 1989). Eskeland (1981) also observed inactivity and dozing which extended to 56 weeks after surgery. Further evidence for pain-related guarding behaviour comes from a study where the birds were presented with drinking water ranging in temperature from 20 to 45°C (Gentle *et al.*, 1990). Partial amputation of the beak resulted in significant behavioural changes with reductions in environmental pecking, beak wiping and head shaking. Pecking at the water presented at 45°C, and drinking at all temperatures, were also reduced after amputation. These behavioural changes have been interpreted as instances of guarding behaviour and hyperalgesia which persists for at least 6 weeks, 3 weeks after the beak had healed. Wall (1979) has suggested that there are several phases in response to painful injuries in animals. The immediate phase, which is often painless, is followed by the acute phase which is characterised by a combination of tissue damage, pain and anxiety. This acute phase is followed by healing and recovery or it leads to the chronic pain phase which is characterised by increased sleep, inactivity and disturbances of eating, grooming and social behaviour. Many of the behavioural changes associated with beak trimming in adult birds could result from a chronically painful condition and depression (Fraser and Quine, 1989).

In the days that following amputation electrical recordings from the nerve fibres in the stump of the beak show features not seen in normal trigeminal afferent fibres (Breward and Gentle, 1985). The most characteristic abnormality encountered in the beak stump was the presence of large numbers of spontaneously active nerve fibres with either regular, irregular or bursting discharge patterns. These abnormal responses were recorded from the beak stump at 5 to 83 days after initial amputation and the receptive fields of these fibres were located on the distal tip of the stump and at varying distances (up to 12 mm) proximal to it. This spontaneous activity seen in the amputated stump was similar to that observed in the experimental neuroma preparations developed initially by Devor, Wall and co-workers (Devor and Bernstein, 1982; Govrin-Lippmann and Devor, 1978; Wall and Gutnick, 1974) in the rat and later extended to the mouse (Scadding, 1981) and cat (Blumberg and Janig, 1984).

Provided the amount of tissue removed is moderate, beak trimming in chicks does not result in neuroma formation that persists to adulthood. Severely trimmed beaks did produce neuroma which persisted in mature hens (Lunam *et al.*, 1996). In an anatomical study of the nerves in the beak of 5 week old brown leghorn birds from 1 hour to 70 days after amputation (Gentle, 1986), it was found that the beak had a limited ability to regenerate normal beak structure in these older birds. By 15 to 30 days after amputation the stump had healed. The beak then continued to grow but the normal dermal structure did not regenerate. The healed stump contained a continuous layer of epithelium with outer keratin sheath overlying an extensive area of scar tissue. Adjacent to the scar tissue the damaged and regenerating nerve fibres formed extensive neuromas and it is likely that these neuromas give rise to the abnormal neural activity.

Studies on peripheral nerve injury and subsequent neuroma formation in mammals have suggested that abnormal activity arising from regenerating axons are implicated in post-amputation stump pain (Seltzer *et al.*, 1991; Wall, 1981) and similar conditions would apply to beak trimming in the chicken. Although neuromas have been identified in the beak together with abnormal neural activity it does not follow that all birds will suffer long term chronic pain.

In human patients stump and phantom pain is reported to occur in about 70% of patients within the first 2 years after amputation (Jensen *et al.*, 1983,1985). The pain is generally believed to fade away and finally disappear except in 5 to 10% of patients where pain persists and may even worsen with time (Melzack, 1971). If similar conditions apply to the chicken then we might expect the same proportions of animals to suffer pain as a result of beak trimming.

(c) Sensory deprivation

The beak of both the chicken and turkey receives an extensive nerve supply from the trigeminal nerve and contains large numbers of sensory receptors which are important for touch, temperature and nociceptor sensitivity. In the lower beak of the chicken there are 15-20 dermal papillae which extend into the thick keratin of the exterior of the beak and contain large numbers of encapsulated sensory endings and free nerve endings (Gentle and Breward, 1986), forming a specialised beak tip organ. The upper beak does not have these sensory papillae but the dermis at the tip of the upper beak contains very large numbers of sensory endings. The large number of receptors at the tip provides the animal with a higher resolution of tactile sensory information from the beak tip and enables the animal to perform the complex manipulative tasks associated with the beak. Beak trimming removes this sensitive area of the beak and although significant regrowth of the beak occurs in young birds the regenerated beaks contain fewer sensory corpuscles (Dubbeldam *et al.*, 1995; Lunam *et al.*, 1996).

This altered sensory perception may impair the ability to pick up food; the impairment is especially marked in the case of particulate diets (Gentle *et al.*, 1982) to the extent that, if too much beak is removed and the birds have access only to pellets, it can result in greatly increased mortality (Deaton *et al.*, 1987). It has been shown that beak trimming adult hens (Gentle *et al.*, 1982) reduced feeding efficiency (number of pecks per gram of pellets ingested) to only 20% of its pre-operative value. Frame-by-frame analysis of cine film showed that the bird was either failing to grasp the pellet in the beak, or not transferring it to the pharynx where it could be swallowed. The adult bird could not adapt its stereotyped behaviour pattern to compensate for the altered beak shape. The re-shaped beak following beak trimming is also inappropriate for feeding in young chicks. Reduced feeding efficiency was seen in beak trimmed chicks; non-trimmed birds were observed to swallow a significantly higher proportion of the seed (Workman and Rogers, 1990). Hogan (1973) has proposed that the tactile feedback generated from pecking seeds forms a reward system which develops during the first 2 days of life. During the first week of post-hatching life it is likely that the mandibulation and ingestion of food items constitute two separate feedback reward systems, the post-ingestional reward systems not developing until at least 4 days after hatching. This proposal by Hogan was taken further by Workman and Rogers (1990) who suggested early pecking preferences are influenced by the reward gained from tactile cues supplied from the beak regardless of whether the food is swallowed. Beak-trimming appears to increase the importance of feedback from post-ingestional factors of nutrition relative to that of tactile input. These findings may have some relationship to "starving-out" (i.e. chicks which starve because of an inability, or refusal, to eat during the first 3-6 days of post-hatching life). The period when starve-outs are most common occurs during the critical period of food learning. This data would suggest that beak trimming may reduce food intake in young chicks and so increase the number which die through starvation or, at least, are underweight.

IV. MECHANISMS OF ACTION OF BEAK TRIMMING

In general the poultry industry considers beak trimming to be an important procedure for the control of feather pecking and cannibalism especially in laying hens which are not kept in

battery cages. Although beak trimming is widespread there is evidence that trimming is not always completely effective (Hughes and Michie, 1982; Denbow *et al.*, 1984; Leighton *et al.*, 1985). In the UK growing broilers are not trimmed and neither are growing turkeys although it is necessary to trim breeding turkeys because of the high light intensities required for photostimulation to bring them into breeding condition. Breeding turkeys which are not beak trimmed are at a considerable risk from cannibalism (Grigor *et al.*, 1995).

The situation varies considerably for other species. For some species of duck, such as Muscovies, beak trimming is essential if they are to be kept under intensive conditions, whereas in others, such as Pekins, it is not necessary (Rauch *et al.*, 1993). Guinea fowl kept intensively and beak trimmed show a reduction in food intake and growth rate with no concomitant economic benefits (Oguntona *et al.*, 1988), whereas in the case of pheasants it is essential either to beak trim them or take other steps to prevent damaging pecking, such as fitting bridles (Faure *et al.*, 1993).

At present we do not know why beak trimming is effective and the reasons may differ depending on the age at which the birds are trimmed. Two hypotheses have been considered for beak trimming young birds (Hughes and Gentle, 1995).

1. Trimming shortens the beak making accurate pecking difficult. Pecking becomes less rewarding and this results in a stable, learned inhibition of pecking.
2. Failure to fully regenerate the sensory innervation of the beak results in incomplete sensory feedback which reduces pecking. There is no evidence of pain resulting from beak trimming young birds but in older animals the possibility of pain is very much greater so in older animals there is a third hypothesis.
3. Chronic pain state originates from the amputated stump which reduces pecking.

It seems possible that following beak trimming of older birds, one if not all of these hypotheses would apply. In the chick however there is no evidence of pain being a factor and beak regrowth is so rapid and effective that in a short period of time after trimming the birds are physically capable of feather pecking (Grigor *et al.*, 1995; Gentle *et al.*, 1995; Gentle *et al.*, 1997). A study by Hughes and Michie (1982) concluded that it was the act of beak trimming itself which had the effect and subsequent beak regrowth was irrelevant. In the absence of any further experimental evidence all we can say is that either hypothesis or both may be correct.

V. CONCLUSIONS

It is recognised that feather pecking is influenced by a number of different factors including light intensity and type of housing, social factors such as group size and stocking density (Hughes and Duncan, 1972; Blockhuis, 1989), genetic factors (Cuthbertson, 1980) and a variety of other different motivational factors (Allen and Perry, 1975; Savory, 1994). Given the variety of factors involved it is not surprising that feather pecking and cannibalism have been very difficult to control without beak trimming. If beak trimming is necessary then there are a number of factors which need to be considered. The first is the age at which the birds should be beak trimmed. From our research beak trimming of chicks does not give rise to any long-term painful consequences and therefore it would be best limited to chicks. At present it is not possible to recommend the best age. It seems that fewer complications are likely to occur if the birds are trimmed after they have learnt to feed and in the UK most turkey producers trim at either 6 or 21 days of age. In the chicken a number of commercial producers tend to trim at 10 days of age although trimming at the hatchery is still common. The second factor is the amount of beak which should be removed. For the general welfare of the bird the least amount of beak which will control cannibalism should be removed. This will depend on the strain of the bird

and the rearing conditions but the complications resulting from severely trimmed birds reported by Lunam *et al.* (1996) should be avoided. It is also important to remove enough beak to prevent the need to trim a second time at an older age. In the UK the aim is to remove approximately a third of the upper beak of the turkey which can be reliably performed with secateurs. In a series of experiments (Grigor *et al.*, 1995) we found that using a hot blade debeaker or the Bio-beaker removed a variable amount of beak but considerably more of the beak than with the secateurs. The effectiveness at reducing cannibalism in all three methods depended on the amount of beak removed. Therefore, because of the precision of the secateurs method, to control cannibalism effectively in the turkey it might be better to remove slightly more of the beak with secateurs.

Although the adverse effects of beak trimming chicks is relatively minor it is a traumatic procedure which deprives the bird of an important source of sensory information. It has been known that for many years that the incidence of feather pecking and cannibalistic pecking differs between different breeds and stock of domestic fowl (Hughes and Duncan, 1972; Robinson, 1979; Craig and Lee, 1990; Blokhuis and Beuving, 1993) and in future poultry geneticists need to incorporate behavioural/welfare traits in their selection programmes to prevent the need to beak trim birds.

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MEASUREMENTS OF FEEDING VALUE OF WHEAT AND BARLEY FOR BROILERS

T.A. SCOTT¹, H.L. CLASSEN², M.L. SWIFT³, G.G. IRISH²Summary

The main objective of this research is to develop a sensitive bioassay, capable of accurately measuring variability between cereal grains or by-products, and the effect of feed additives and processing. Data and samples from 108 wheat and 97 barley samples tested in the bioassay have been used to validate lab-bench (i.e. carbohydrate profiling and near infra-red spectrometry) predictors of feeding value.

The apparent metabolisable energy (AME) bioassay has evolved into a sensitive measurement of feeding value of cereal grains for broilers. Over time, and many samples, it has been possible to reduce the number of collection times, the number of replicate pens used for each test diet, and the necessity to do many tedious measurements of gross energy (GE) to determine ME. Using the broiler bioassay, large genetic and environmental variations in AME, growth and efficiency have been found among diets containing 800 g/kg of a test wheat or barley with or without an enzyme. This variation in feeding value can be accurately predicted with near infra-red spectrometry (NIRS) of whole grain samples, and to a lesser extent with measurements of extract viscosity and select non-starch polysaccharides (NSP).

I. INTRODUCTION

The Agassiz AME bioassay has a striking resemblance to that used by Sydney University (Mollah *et al.*, 1983; Rogel *et al.*, 1987). The present involvement in measuring feeding value of cereals, and the development of the Agassiz AME procedure, grew out of discussions with members of the British Columbia feed/nutrition council. Their concerns were: a) what is the feeding value of the new feed (CPS white) wheats (Canadian Prairie Spring); b) why don't the TME measures of the cereals fed relate to broiler performance in the field; and c) how much do feed enzymes in wheat- and barley-based diets improve the digestibility of energy and amino acids. Ways of measuring AME were discussed with funding to support an effort to use broilers to measure feeding value of wheat and barley.

II. MATERIALS AND METHODS

(a) AME procedure

The AME procedure has been fully described (Scott *et al.*, 1997a). Briefly, 50 kg of test cereal are required. Upon receipt, a subsample of whole grain is taken and used to determine bushel and 1000 kernel weight, then stored for future reference. Approximately 45 kg of the whole grain is finely ground, subsampled, and then mixed (at 800 g/kg of diet) with a basal supplement (Table 1) containing 11 g/kg acid insoluble ash as a digestibility marker.

¹ Agriculture and Agri-Food Canada, PO Box 1000, Agassiz, BC, V0M 1A0, Canada.

² University of Saskatchewan, Saskatoon, SK, S7N 5B5, Canada.

³ Pro Form Feeds Ltd, PO Box 1000, Chilliwack, BC, V2P 6J6, Canada.

The diet is then split, one portion fed *as is* and the other portion supplemented with 1.5 g/kg of an appropriate NSP enzyme¹. Diets with or without enzyme are then fed to four pens of male broiler chicks, *ad libitum*, from 4 to 17 d of age. Excreta are collected for 24 h at 8 and 16 d of age. At 17 d the chicks are humanely killed, intestinal tract removed, digesta viscosity measured from digesta of the upper tract and ileal digesta collected and pooled. Diet, excreta or ileal digesta are measured for dry matter (DM), insoluble ash, nitrogen and gross energy. Growth and feed intake of the broilers from each of the four pens are measured during the feeding period (4 to 17 d). Based on analysis of a large number of samples, modifications to this procedure will be discussed briefly.

Table 1. The ingredient profile of a test diet, and typical units in g/kg determined levels of crude protein, starch and extract viscosity for different classes of wheat and barley.

Ingredient in diet	g/kg	Composition of Grains	Wheat 1993			Barley 1993	
			CPS	Durum	HRS	Hulled	Hulless
Test cereal	800.0	Crude protein (g/kg)	125.2	142.1	149.4	126.9	137.4
Tallow	20.0	Starch (g/kg)	643.4	632.3	613.2	582.0	613.1
Isolated soy protein	115.1	Extract viscosity (cps)	1.49	1.19	1.83	4.85	16.53
Corn gluten meal	11.0		Wheat 1994			Barley 1994	
L-Lysine	1.5		CPS	Durum	HRS	Hulled	Hulless
DL-Methionine	0.40	Crude protein (g/kg)	151.2	165.2	177.7	128.8	156.6
Vitamin/mineral premix	41.0	Starch (g/kg)	644.3	631.0	631.4	582.0	613.6
Celite (marker)	11.0	Extract viscosity (cps)	1.57	1.22	1.95	4.89	11.99

(b) Samples measured

A total of 108 samples of wheat (nine cultivars grown in duplicate in three locations in each of two crop years) and 71 samples of barley (14 cultivars grown in several locations over three crop years) were analysed using the broiler chick bioassay described by Scott *et al.* (1997b). The wheat samples included three Hard Red Spring (HRS; CDC Teal, Katepawa, Laura), three Durum (DUR; Kyle, Plenty, Sceptre), and three Canadian Prairie Spring (CPS; Biggar, Genesis, Glenlea) cultivars. Durum wheat cultivars, commonly used for pasta, are a different species (tetraploid) as compared to the HRS and CPS wheat (hexaploid). In Canada, classes of wheat must be distinguishable by kernel type. However, they are also identified by gliadin banding with electrophoresis. These nine cultivars were each grown in test plots in Saskatchewan and Alberta in duplicate in each of two crop years (1993 and 1994).

¹ FinnFeeds International, Box 777, Marlborough, Wiltshire, United Kingdom, SN8 1XN.

The 14 barley cultivars included four hulled and 10 hullless cultivars. Of the 14 barley cultivars, five were six-row and nine were two-row, one was a waxy cultivar and two were malting cultivars. Growing locations were distributed over western Canada and varied between the three crop years.

(c) Lab-bench predictors

The data and samples of whole grain, ground grain, diet, excreta and ileal digesta were made available to H. L. Classen (University of Saskatchewan) and M.L. Swift (Pro-Form Feeds Ltd) to validate, respectively, the use of chemical analysis and Near Infra-Red Spectrometry (NIRS) as lab-bench predictors of cereal grain feeding value.

For wheat, the different samples were analysed for starch, protein, ether extract, neutral detergent fibre, acid detergent fibre, dietary fibre and extract viscosity (Bedford and Classen, 1993). As well, levels of soluble and total NSP and, specifically soluble and total pentosans were measured (Englyst and Hudson, 1987; Englyst, 1989). These values were correlated with measurements from the AME bioassay.

NIRS of whole and ground grain and test diets were conducted by M.L. Swift. Based on spectral analysis of 128 whole wheat, test diet and excreta samples, calibrations were made for approximately 50 bioassay and chemical measurements.

III. RESULTS AND DISCUSSION

(a) Evolution of AME procedure

The AME bioassay procedure relies on diet and excreta (or ileal digesta) measurements of DM, GE and level of insoluble ash. In particular, GE measurements of diet and excreta are time consuming and expensive laboratory operations. Two predictor equations were tested: one did not require GE measurements of excreta or digesta, and the other eliminated the necessity of doing GE of the diet as well (Figure 1). The results showed that GE measurements of excreta and digesta could be eliminated if a prediction error of 80 kcal/kg (<3%) or less for ME could be accepted. Development of a more sensitive assay for measuring the "natural" levels of acid insoluble ash of commercial diets, i.e. no added marker, may prove to be a valuable tool for feed companies to screen diets.

Based on data from 276 wheat and 194 barley diets, AME was determined using excreta collected at 16 d using data from four, three or two replicate pens of six male broilers, and values correlated as an assessment of accuracy with fewer replicates. Overall, the mean values determined with three and two pens as compared to four pens were highly correlated ($r^2 = 0.99$ and 0.97 , respectively). Furthermore, it was observed that variability among pens of birds decreased as the broilers became older (i.e., 16 vs 8 d of age). The AME measurements were also significantly higher when determined at 16 d as compared to 8 d, the differences being approximately 1.5 and 5.0% for barley and wheat, respectively.

A 3% difference in AME due to grain type, enzyme addition or processing treatment can be determined with this modified AME procedure. This procedure would not require GE determinations of excreta, and would only require two or three replicate pens of six male broilers as compared to the four pens currently used.

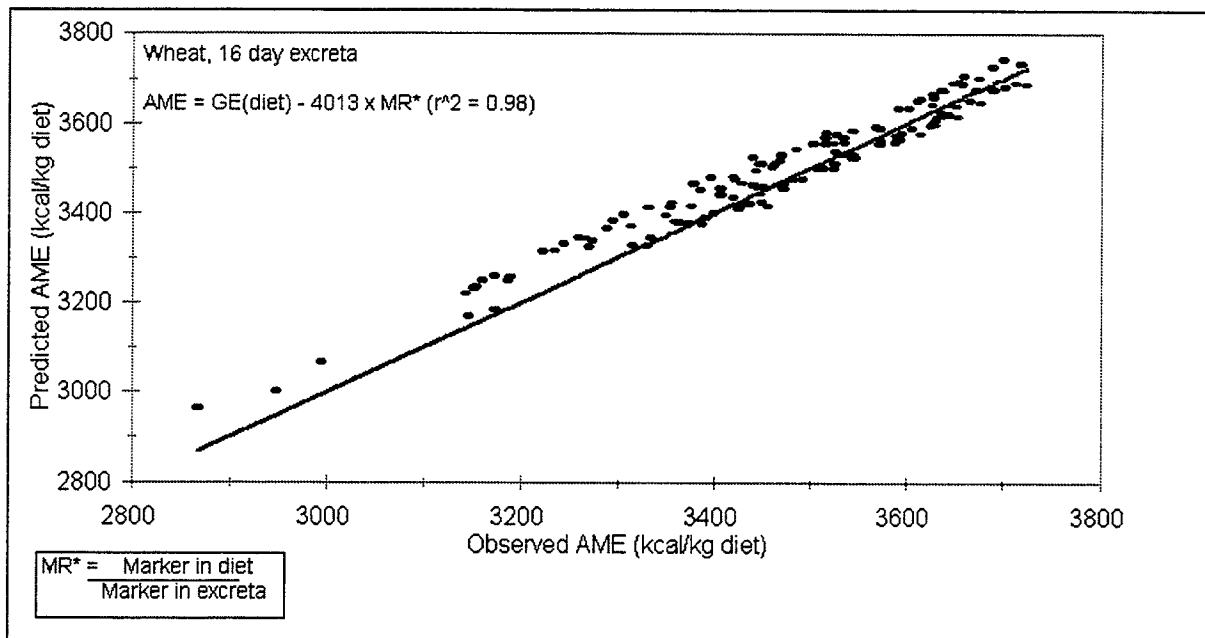


Figure 1. The AME (kcal/kg diet) based on 16 d excreta collection (24 h) using predictor equations with GE measurements of diet and without GE measurements of excreta. (Without diet GE: $AME = 4604 - 3991 \times MR (r^2=0.94)$).

(b) Variability in AME of wheat and barley

The AME of samples of wheat and barley provided by cereal geneticists from western Canada over two crop years were measured. The samples were of good quality and treated agronomically in a controlled manner, although from different geographical locations and subjected to different weather conditions. The mean values in Table 2 represent the average AME of the four individual pens of six broilers used to assess each cereal sample with or without enzyme. Therefore, the values provided as the minimum or maximum are indicative of variation due to sample source and individual pen variation.

For wheat-based diets, the AME of Durum wheat was significantly higher than those for CPS and HRS; the latter classes of wheat were not significantly different, in both crop years and with or without enzyme supplementation. Although Durum wheat is used by the pasta industry, inclement harvesting conditions often make it available as a feed wheat. Based on the feed industry's perception of the feeding value of Durum wheat it had been avoided and had to be "mixed-off" at a maximum inclusion of 10% with other classes of feed wheat. So, these results indicate that a review of this policy for grain handlers should be revisited. The other surprising response was the overall good performance of the CPS, as compared to the HRS, wheat as again CPS (feed wheat) has been "suspect" by the feed industry. Comparison of individual cultivars of CPS wheat support this suspicion by the industry. One cultivar (Glenlea) was nearly equal in feeding value to the Durum wheat cultivars and much higher than the other two CPS wheat cultivars.

Table 2. Apparent metabolisable energy (AME) (kcal/kg diet) determined from 24 h excreta collection from four individual pens of six broilers per diet at 16 d of age.

	1993 Crop Year									
	With Enzyme			Without enzyme			With Enzyme		Without Enzyme	
	CPS	Durum	HRS	CPS	Durum	HRS	Hulled	Hulless	Hulled	Hulless
n	72	72	72	72	72	72	12	64	12	64
Mean	3740 ^b	3850 ^a	3670 ^b	3130 ^b	3510 ^a	3210 ^b	3410 ^b	3510 ^a	3080 ^a	3080 ^a
SD	115	69	135	254	60	201	42	111	94	214
Min	3320	3650	3130	2270	3200	2770	3320	3290	2960	2470
Max	3950	3960	3890	3660	3840	3660	3470	3710	3210	3560
	1994 Crop Year									
n	72	72	72	72	72	72	80	180	80	180
Mean	3610 ^b	3710 ^a	3610 ^b	3480 ^b	3630 ^a	3480 ^b	3200 ^b	3350 ^a	3040 ^a	3000 ^a
SD	85	54	103	147	65	146	78	105	125	199
Min	3380	3580	3260	3030	3470	3100	3030	3080	2700	2540
Max	3790	3810	3780	3720	3760	3740	3370	3590	3330	3390

^{ab} Mean values with different superscripts within crop year and enzyme supplementation categories are significantly ($P < 0.05$) different.

For barley, there were no significant differences between hulled and hulless barley classes when the diets did not contain enzymes, as has been previously reported by Campbell and Bedford (1992). With enzyme supplementation, the hulless as compared to hulled barley consistently had a higher feeding value and resulted in significantly better broiler performance. Within the individual cultivars of hulless barley, two cultivars (SB90300 and Falcon) had superior AME values. Based on these results on-going research is being conducted to measure the feeding value of new low-fibre, hulless barley cultivars.

(c) Chemical and physical prediction of feeding value of wheat

Development of lab-bench predictors of broiler performance on wheat-based diets has been evaluated, based on selected samples of wheat differing in cultivar and growing location. Prediction of the AME levels of wheat-based diets with or without enzymes are presented in Table 3. Overall, there were generally higher correlations between measures of feeding value and physical and chemical measurements when diets were fed without enzyme. This reflected the higher variability between diets without enzymes as indicated in Table 2.

Table 3. The correlation coefficients between bioassay measurements (AME (kcal/kg)), bird performance (feed intake (FI; g/bird/d), body weight (BW; g) and feed conversion (F:G, g:g)) and digesta viscosity (Visc; cps), and chemical and physical measurements of wheat and wheat-based diet. (Only significant r^2 ($P < 0.05$) values are reported).

Variable	Wheat-based diets with enzyme					Wheat-based diets without enzyme				
	AM E	FI	BW	F:G	Visc	AME	FI	BW	F:G	Visc
BW @ 17 d	-0.20	0.99	1.00	-0.85	0.39	0.37	0.99	1.00	-0.90	-0.19
Feed:Gain (F:G)	--	-0.79	-0.85	1.00	-0.22	-0.53	-0.83	-0.90	1.00	0.34
Digesta viscosity	-0.39	0.46	0.39	-0.22	1.00	-0.67	--	-0.19	0.37	1.00
%Protein wheat	-0.17	0.59	0.64	-0.65	--	0.28	0.58	0.63	-0.66	--
%Starch wheat	--	0.23	0.21	--	0.28	--	0.24	0.21	--	--
Extract visc	-0.50	--	--	--	0.33	-0.57	--	--	0.20	0.82
Soluble pentosans	-0.40	-0.24	-0.27	0.30	0.21	-0.67	-0.30	-0.36	0.50	0.61
Total pentosans	-0.44	-0.33	-0.39	0.46	--	-0.62	-0.42	-0.49	0.61	0.61
Soluble NSP	-0.32	-0.20	-0.23	0.26	--	-0.59	-0.27	-0.33	0.47	0.54
Total NSP	-0.41	-0.19	-0.23	0.34	--	-0.47	-0.28	-0.34	0.45	0.53
1000 Kernel Wt	0.51	--	--	--	-0.23	0.58	--	--	-0.24	-0.69

Considering that wheat-based diets are routinely fed, in most cases, with an enzyme, it would appear that measurements of the carbohydrate profile of the cereal grains would not relate well to measurements of AME or broiler performance. Similarly, it is even more disappointing that the AME of wheat-based diets do not correlate well with feed conversion measurements, which would be expected if the birds' intake was inversely proportional to the AME content of the diet. The relationship between AME and feed:gain ratios was -0.53 for wheat-based diets without enzyme, reflecting the higher variability among these samples as compared to when enzymes were included. Data from the present samples of wheat did not verify the strong relationship reported by Annison (1993) between wheat AME and the concentration of water soluble pentosans ($r^2=0.81$) and soluble NSP ($r^2=0.90$).

The other observation (data not included here) was that correlations between soluble NSP measurements of grain were highest with AME determined at 16 d as compared to 8 d of age. This is unexpected if young birds are more susceptible to viscous carbohydrates than older birds. It does, however, support observations that the broiler response to enzyme supplementation of wheat-based diets is higher in older, than in younger, birds.

(d) Near infra-red spectrometry prediction of feeding value of wheat

To date, the NIRS study has focused on using the spectra from whole wheat samples only. From casual observation of the spectra, the classes of wheat have distinctive spectra, which should help in identifying wheat cultivars. Furthermore, NIRS calibrations were developed for AME, extract viscosity, soluble arabinoxylans, total arabinoxylans, kernel

weight and measurements of bird performance. Based on these calibrations we were able to calculate the correlation between predicted and actual values (Table 4).

Table 4. Statistics for calibration equations developed using near infra-red spectrometry (NIRS) whole wheat scans.

Parameter	Mean	R ²	SECV ¹
AME diets without enzyme (kcal/kg)	3530	0.93	56.90
AME diets with enzyme (kcal/kg)	3670	0.80	65.50
Extract viscosity (cps)	1.52	0.93	0.12
Soluble arabinoxylans %	1.61	0.84	0.24
Total arabinoxylans %	6.07	0.94	0.24
1000 Kernel weight (g)	40.39	0.95	1.55
Body weight (g) at 17 d	364	0.93	31.69
Feed:Gain (g:g) 4 to 17 d	1.54	0.97	0.08

¹SECV = Standard Error of Cross Validation

Of significance are the high correlations between predicted measurements for feeding value using NIRS and determined measurements from the bioassay and chemical analysis. Of particular significance are the observations that NIRS can predict animal performance. To the authors' knowledge, similar calibrations have not been available previously. Similar calibrations are currently being developed for the barley samples described earlier.

Future plans for NIRS calibration include its use in predicting the insoluble ash levels of research diets and diets based on natural levels of acid insoluble ash. In the latter case this information, based on the "new" method of determining AME described above would be a highly valuable tool for predicting field performance of broilers fed commercial diets.

IV. CONCLUSIONS

The evolution of the AME procedure has met the criteria for accuracy and repeatability. Further, based on large data sets, "short cuts" have been developed that will facilitate the handling of greater numbers of samples with minimum requirements for time-consuming measurements of gross energy. Significant variability in feeding value between wheat and barley cultivars were measured. These data were used to validate lab-bench measurements that may prove to be useful in predicting the AME of cereals, and, in the case of NIRS, in predicting broiler performance with diets containing the tested cereal.

The present data simultaneously determined AME and broiler performance. Unfortunately, the relationship between these parameters were not strong, particularly when diets were supplemented with enzymes. Not having a strong relationship between AME and feed conversion could relate to a number of factors including: diet palatability, balance of nutrients in the test diet; gut micro-flora development; and other anti-nutritive factors that impact digestion, absorption and, potentially, metabolic energy requirements.

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THE BIRTH OF THE FORMULA FEED INDUSTRY IN AUSTRALIA 1954-1962

J. DARLING

In 1954 the Poultry industry in Australia lagged well behind the rest of the world in both its breeding and feeding strategies. The Department of Agriculture in New South Wales (NSW) saw no reason to introduce formula feeds. Their then "expert" said that poultry farmers should feed their flocks wet mash in the morning and grain (wheat) in the afternoon with access to clover to graze. There were no cages for layers, no broilers and/or broiler sheds.

The laying hens were mainly White Leghorns with some New Hampshires and Australorps. The male chick of the laying breed was the sole bird available for the so-called "Roast Chicken on Sundays". Hormone implants were just starting to be used to produce the capon. Feed conversion was poor, egg laying percentage was poor and the average Australian ate 1 kg of chicken meat a year. At that time the consumption in the USA was 18 kg per person per year.

The ingredients for the diets of poultry came from the flour millers and from the abattoirs, i.e. bran, pollard and meat meal. Traditionally these were handled by produce merchants in hessian or jute sacks. Bulk feeds were totally unknown. The poultry farms were small, situated on the outskirts of the state capital cities with the possible exceptions of Tamworth in NSW and Bendigo in Victoria.

Being from a flour milling and grain family my first brush with possible improvements was when I worked for experience in Cheadle Heath near Manchester in England with the firm of Henry Simon Ltd, an engineering firm specialising in the design of manufacturing machinery for flour mills and just emerging machinery for provender mills as feed mills were then called in the UK. The other countries that had not dissimilar engineering firms were Switzerland, Germany and the USA.

So it was with youthful expectations that a pilot plant was put together in the flour mill of John Darling and Son at Rhodes in Sydney, and in November 1954 I remember packing the first bag of Dairy Meal. This was rapidly followed by Layer All-Mash, a complete feed formulated in accordance with the growing science of nutrition that was developing apace overseas.

The dairymen wanted the product but the poultry industry was sceptical. However, like most things in life it only needed two or three people to try and embrace the concept and practice for formula feeds to catch on, in particular when in those days it was difficult to get the ingredients from the manufacturers. A few farmers were starting to build cage plants and the cage manufacturers like Multiplo Ltd were active in such promotions. Naturally, the owners of these plants were quick to realise that feed millers could formulate, mix and deliver more cheaply than they could do themselves.

So in 1955 and 1956 egg and dairy producers began to improve their productivity and profits. We imported a pelleting machine from the California Press Company and so a range of pelleted feeds and crumbles for chicks was launched.

About this time one or two growers were building broiler sheds, based on United States design and technology. One of the first was Tom Lewis who had returned to Australia after working in the Australian Embassy in Washington.

"Lakehead", Burrawang, NSW 2577.

He bought a property on the Nepean River at Castlereagh near Penrith and became the first real broiler grower in Australia to utilise deep litter. He later became a politician and the Premier of NSW.

We now had the capacity, the equipment and the nutritional technology to service the poultry, dairy, pig and cattle industries. Minor users were rabbit growers, sheep and horse owners and zoos. Yet in the poultry industry something was lacking - the bird! The old White Leghorn was just not good enough.

So the poultry breeders got to work. Jack Ingham went to the US in 1956 as the leader of the breeders, with names such as Cooper, Druce, Donnelly, Locket and Hazlett all breeding and improving their stock. For the first time in Australia the broiler had arrived as a distinctive breed. Parent and grandparent specialised farms appeared. The feed conversion factor has come down from 4.5:1 to 1.8:1. Consumption per head has now reached more than 27 kg per person and the price of chicken in the marketplace has beaten inflation by a mile and is now the most economic form of animal protein for human consumption. In those days, it was only the occasional rare treat for families to have a roast chicken for Sunday lunch - now available to all as whole chicken or pieces.

In 1955 and 1956 Professor Bouffleur of the Royal Agricultural College in Cirencester, England visited Australia. His speciality and his message was the feeding of concentrates to dairy cattle to lift milk yields and to increase profits. There developed, particularly on the south coast of NSW as far as Milton, the southernmost limit of the milk zone, an amazing increase in the feeding of dairy meal and dairy cubes formulated to Professor Bouffleur's specifications. This, of course, has continued ever since, now spreading throughout Australia. Interestingly, as formula feeds were developed, the breeders of dairy cattle saw the advantages that the Friesian and Holstein breeds of cattle brought to the production of whole milk. The Jersey and Guernsey breeds, in particular, could not compete for the growing milk market resulting from the expansion of the cities. In NSW they remained for some considerable time in the north around Lismore and Casino and in the south around Bega and Kameruka supplying the need for cream and cheese products.

A similar pattern was developing in the other states. Many producers with Ayrshire and Illawarra Shorthorns gradually changed to Friesians until today it is rare to see any cattle other than Friesian/Holstein in the dairy country of Australia.

It was not until the advent of formula feeds and, particularly, pellets and cubes that the pig industry emerged from the days when the poorest of farmers ran a few pigs fed on swill and waste from bakeries, vegetable scraps and spoilt grain.

Gradually pig farms became larger, and feeds were designed for various ages of pigs. Bulk deliveries made the task of feeding easier and more efficient so that today it is rare to find a farmer running a few pigs in third rate circumstances. The large pig operations are now very large, scrupulously hygienic and the quality of meat enhanced by the improvement of feeds.

We must also acknowledge that until the ability to secure pelleted or cubed feeds was achieved the cattle feed lot did not develop. The last twenty years has given birth to the production of beef quite separately to the continuing and traditional production of grass-fed cattle.

The American and, latterly, Japanese and Korean demand for marbled and tender meat has created a new revolution in the supply side. Very large feed lots now operate in Australia. Most are foreign owned and vertically integrated, with traditional breeders in part changing to newer breeds more suited to the feed lot environment. So many new breeds are appearing. The Shorthorn breed once predominant in the North of Australia is a diminishing breed as is the Hereford in the South. The two breeds that have increased in importance are the Angus

and the Murray Grey with the black Angus particularly so. This has been encouraged by the Japanese paying a premium for this meat.

From 1954 to 1962 the formula feed industry grew apace with other mills being started, and mills such as Darlings in NSW, Kimptons and Minifies in Melbourne, and N.B. Love Millmaster in Sydney being predominant.

Acquisition and integration began with millers acquiring wholly or in part breeders and poultry processors. Breeders became larger by acquisition and merger. Large corporations became involved, such as Amatil and George Weston and Allied Mills. The latter are now part of the Goodman Fielder Wattie group. Amatil have now withdrawn from the poultry industry. Steggles in Newcastle became part of the Goodman Fielder Wattie group as had John Darling and Son earlier.

The University of Sydney helped by establishing the Department of Animal Husbandry under Professor Terry Robinson. He obtained financial support from the Australian Dairy Board, the Australian Meat Board and Wool research sources to purchase two farms at Camden to be developed as a major teaching and research centre.

In 1959 both the Poultry Husbandry Research Foundation and the Dairy Husbandry Research Foundation were formally established by the University and in 1962 the World Poultry Science Association held its World Congress in Sydney. This recognition of the industry in Australia acted as a further spur to an industry that modernised itself in line with the best standards in the world.

I like to think that the animal feed manufacturers played a small part in throwing the first stone into the pool that truly allowed all animal and avian industries to play a real part in Australia's growth.

EFFECTS OF HEAT STRESS ON EGG AND EGG SHELL QUALITY IN FIVE STRAINS OF LAYING HEN

J.R. ROBERTS and W. BALL

Summary

Egg and eggshell quality were assessed in five strains of laying hen: the imported strains Isa Brown, Lohmann Brown and Hy-Line Brown and the Australian strains Tegel Tint, and Hy-Line-CB before, during and after a period of heat stress. A control group of birds was maintained at a constant 20°C. The heat-stressed group was maintained initially at 20°C, then subjected to heat stress for two weeks by daily cycling between 33°C and 20°C, followed by a period of constant 20°C. The heat stress resulted in a reduction in feed consumption, reduced egg size, lighter shell colour and poorer egg shell quality as evidenced by shell weight and shell thickness. Shell breaking strength was not significantly affected, suggesting that changes occurred in egg shell ultrastructure. Some strains were more affected than others by the heat stress.

I. INTRODUCTION

Losses to the Australian egg industry, resulting from poor egg shell quality, have been estimated at 10% or more of total egg production. One known cause of egg shell quality problems which occurs commonly in Australia is heat stress caused by high ambient temperatures. The extent of stress will be influenced by factors such as humidity and the extent to which the hens have become acclimatised. The deleterious effects of heat stress on egg shell quality appear to be due to several reasons. Feed intake is usually depressed (Marsden and Morris, 1987) and this may result in a decreased calcium consumption. Also, at high temperatures, birds pant to enhance evaporative cooling. Panting results in respiratory alkalosis which is caused by loss of CO₂ from the blood and involves an increase in blood pH (Mongin, 1978). This, in turn, decreases the proportion of the blood calcium that is in the ionised form and thus reduces the amount of calcium which is available for egg shell formation. The activity of carbonic anhydrase (the enzyme which produces bicarbonate for shell formation) may also be reduced during heat stress and blood flow to the uterus (shell gland) may decrease.

The Australian commercial layer industry utilises a range of strains including Australian bred strains, imported strains and hybrids between the two. The susceptibility to heat stress varies among strains (Arad *et al.*, 1975) and it is reasonable to assume that some strains may be better suited to high ambient temperatures.

The present study compared the effect of heat stress on egg and egg shell quality in three imported strains and two Australian strains of commercial laying hen. A cyclic elevated temperature was used to simulate natural conditions.

Division of Animal Physiology, School of Rural Science and Natural Resources, Faculty of the Sciences, University of New England, Armidale, NSW 2351.

II. METHODS

Twelve birds of each of the imported strains Isa Brown, Lohmann Brown, Hyline Brown and the Australian strains Tegel Tint and Hy-Line-CB were housed individually in cages in constant temperature rooms in the Animal House at the University of New England. The birds had been transported as day-olds from their respective hatcheries and reared together in the Animal House. Birds were transferred to a commercial-style layer shed prior to the onset of lay and maintained on a commercial ration. The heat stress experiment was conducted when the birds were 45 weeks of age. Six birds of each strain were housed in a constant temperature room which was maintained at 20°C for the duration of the experiment (control group). The other six birds of each strain were housed in a separate room which was maintained at 20°C except for the two week period of heat stress (heat-stress group). All birds were allowed a period of 5 weeks to acclimate to the constant temperature rooms. Following this acclimation period, all eggs laid were collected daily for a period of two weeks. The temperature of the room housing the heat-stress group was changed to a cycling 33°C during the day (0060 to 1800) and 20°C during the night (1800 to 0060). All eggs laid were collected during this two week period. The temperature of the heat-stress room then reverted to a constant 20°C for a further two week period during which all eggs laid were collected. The following measurements were made on the eggs: egg weight, gross egg shell defects, egg shell pigmentation (by reflectivity), egg length and breadth, eggshell breaking strength (by quasi-static compression), shell weight and shell thickness (using a dial comparator gauge). Shape index (breadthx100/length) and percentage shell (shell weightx100/egg weight) were calculated. Internal egg quality was assessed by measuring the Haugh Units of the albumen and the Roche score of the yolk. Daily feed consumption was measured for individual birds throughout the experiment. Data were analysed by two factor ANOVA. Fisher's (Protected) Least Significance Difference test was used to determine differences between means. Significance was assumed at $P < 0.05$.

III. RESULTS

Mean body weight was not significantly different between groups at 2038 ± 36 g for the control group and 2069 ± 36 for the heat-stress group. The results for feed consumption are summarised in Table 1. In the control group, feed intake was lowest during the first two weeks of the experiment and increased to a stable level during the subsequent four weeks. For the heat-stressed group, feed intake was depressed significantly by 7% during exposure to heat stress.

Table 1. Feed Intake ($\text{g} \cdot \text{hen}^{-1} \cdot \text{day}^{-1}$) in response to heat stress.

	Before Heat Stress	During Heat Stress	After Heat Stress
Control Group	^b 117.8±1.5	^a 122.1±1.0	^a 123.3±1.0
Experimental Group	^a 121.1±1.4	^b 113.6±0.9	^a 121.8±1.0

Mean±SEM. Values across a row with different superscripts are significantly different from one another.

Results for egg and egg shell quality are summarised in Table 2. Percentage production was not different between the control and heat-stress groups and did not change over the course of the experiment. Percentage production averaged 90% for both groups. Egg weight increased slightly but significantly in the control group over the course of the experiment.

For the heat-stress group, egg weight was significantly lower during the period of heat stress and increased to pre-heat-stress levels when the ambient temperature returned to a constant 20°C. Although all strains exhibited the same pattern, the most marked reductions in egg weight occurred in the Lohmann Brown, Hy-Line Brown and Hy-Line CB birds. Egg length and width both varied in proportion with egg weight such that shape index (egg breadth as a percentage of egg length) was not significantly affected by heat stress.

Egg shell reflectivity (an indicator of shell colour) did not change over the course of the experiment in the control group. However, in the heat-stress group, shell reflectivity was significantly higher during the period of cyclic high ambient temperature, indicating that shell colour was lighter. Although this occurred in all strains, for all except the Hy-Line Brown strain shell colour became darker following the heat stress period. For the Hy-Line Brown birds, shell colour remained lighter after the temperature of the room was returned to a constant 20°C.

Egg shell breaking strength did not change over the course of the experiment in either the control or heat-stress group, although mean shell breaking strength was consistently slightly higher in the control group. Shell weight remained constant in the control group over the course of the experiment but was significantly decreased during the heat stress period in the heat-stress group. Reductions in shell weight in response to heat stress were greatest for the Lohmann Brown, Tegel Tint and Hy-Line Brown birds. Shell thickness did not change over the course of the experiment in the control group. For the heat-stress group, shell thickness was lowest during the period of heat stress due mainly to reduced shell thickness in the Tegel Tint and Hy-Line Brown birds. The percentage shell (shell weight as a percentage of egg weight) was not significantly different for either group.

Table 2. Egg and egg shell quality in response to heat stress

	Group	Before Heat Stress	During Heat Stress	After Heat Stress
% Production (eggs/hen/100 days)	Control	88.8±1.4	91.2±2.1	90.7±1.3
	Heat-stress	90.3±1.7	90.7±1.6	89.2±2.2
Egg Weight g	Control	^b 59.4±0.2	^{ab} 59.9±0.2	^a 60.2±0.2
	Heat-stress	^a 59.7±0.3	^b 58.5±0.3	^a 59.7±0.4
Shape Index %	Control	75.05±.16	75.11±0.15	75.96±0.23
	Heat-stress	73.80±0.35	74.21±0.22	74.16±0.15
Shell Reflectivity %	Control	44.6±0.7	44.8±0.6	44.9±0.6
	Heat-stress	^b 46.7±0.8	^a 48.2±0.8	^b 46.9±0.7
Shell Breaking Strength N	Control	36.2±0.4	36.0±0.3	36.4±0.3
	Heat-stress	34.6±0.4	34.5±0.4	34.1±0.5
Shell Weight g	Control	5.40±0.03	5.45±0.03	5.46±0.03
	Heat-stress	^a 5.35±0.04	^b 5.21±0.03	^a 5.35±0.04
Shell Thickness µm	Control	^b 363.7±1.6	^{ab} 367.3±1.4	^a 368.5±1.7
	Heat-stress	^{ab} 362.1±1.9	^b 358.2±1.5	^a 364.9±1.7
% Shell	Control	9.12±0.04	9.07±0.04	9.12±0.04
	Heat-stress	9.06±0.05	8.96±0.05	8.99±0.05
Haugh Units	Control	90.94±0.4	90.79±0.4	91.23±0.4
	Heat-stress	^b 88.46±0.4	^b 87.97±0.5	^a 89.85±0.4

Mean±SEM. Values across a row with different superscripts are significantly different from one another.

Haugh Units did not change over the course of the experiment in the control group. In the heat-stress group, Haugh Units were lowest before and during the period of heat stress and significantly higher following the heat stress period. Yolk colour score was not affected by heat stress.

IV. DISCUSSION

Relatively few poultry houses in Australia are fully environmentally controlled. Therefore, most laying hens are exposed to high ambient temperatures during the Australian summer. Heat stress, if severe, may result in mortalities. However, less severe heat stress may cause reductions in egg and egg shell quality and associated financial losses. In the present study, five strains of laying hen were subjected to heat stress which was sufficient to induce intermittent panting and postural changes such as holding the wings out from the sides of the body. The intention was to simulate a degree of heat stress which is experienced commonly by commercial laying hens. A cyclic high ambient temperature was selected as being closer to natural conditions than a constant high temperature and also less stressful to the birds. Previous studies have shown that egg production and egg shell quality are less affected by cyclic high temperatures than by constant high temperatures (Daghir, 1995; de Andrade *et al.*, 1977; Wolfenson *et al.*, 1979).

In the present study, feed intake was significantly depressed during heat stress, a finding consistent with other reported studies (see Nys, 1995). Reduced feed intake and associated reductions in calcium consumption would account for some of the reductions in egg shell quality. However, the heat stress imposed in the present study was not sufficient to affect egg production although the eggs laid during the heat stress period were significantly smaller.

The lighter coloured shells of the eggs laid during heat stress may result from a reduction in the amount of pigment deposited in the cuticle or they may be due to the laying down of extra calcium on top of the formed cuticle. The laying down of additional calcium occurs during a range of stressful conditions (Leary *et al.*, 1997) and may reflect a generalised stress response rather than a specific reaction to heat stress.

Shell weight and shell thickness were significantly reduced by heat stress. However, because the eggs produced were smaller, the ratio of shell weight to egg weight did not change significantly. Despite the egg shells being lighter and thinner, egg shell breaking strength was not significantly reduced. A similar finding has been reported previously (Brackpool *et al.*, 1993; Thomas and Roberts, 1995) and appears to result from improvements in egg shell ultrastructure.

The lower Haugh Units occurring during heat stress are likely to be due to more rapid deterioration of the egg albumen at the higher temperature.

Although the present study utilised only small numbers of birds from each strain, some strain differences were found in the effect of heat stress on egg and egg shell quality. This raises the possibility that some strains are better suited to heat stress conditions than others and that this may be one factor influencing the selection of bird strain in some parts of the country.

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RESPONSES OF HEAT-STRESSED BROILERS TO L-ARGININE FREE BASE AND L-ARGININE ACETATE SUPPLEMENTS IN THE DRINKING WATER

J. HAYAT¹, D. BALNAVE¹, J. BRAKE² and M.T. KIDD³

Summary

Supplementation of drinking water with either L-arginine free base or L-arginine acetate failed to improve the weight gain and feed conversion of heat-stressed broilers. Arginine free base had a significantly adverse effect on feed and water intakes and weight gain. Furthermore, a positive response in feed conversion resulting from supplementation of the drinking water with sodium bicarbonate (NaHCO₃) was negated by the simultaneous addition of arginine free base. On the basis of these results arginine supplementation of the drinking water is not a viable alternative to arginine supplementation of the diet for heat-stressed broilers.

I. INTRODUCTION

The importance to broiler performance of varying the dietary arginine:lysine ratio at different temperatures has been highlighted in recent studies (Brake *et al.*, 1994; Gorman *et al.*, 1997). In particular, the liveweight gain and feed conversion of heat-stressed broilers can be significantly improved by widening the dietary arginine:lysine ratio through supplementation with L-arginine free base (Kroon *et al.*, 1997). The current studies were designed to examine whether arginine supplementation of drinking water could elicit a similar response.

II. MATERIALS AND METHODS

Four hundred day-old male broilers fed the same broiler starter diet were grown to 22 (Experiment 1) and 21 (Experiment 2) days of age in electrically heated flat deck brooders in a controlled temperature room maintained at 25°C. Brooder heat was removed at 14 days of age. Birds were randomised on day 21 or 22 into 48 replicates of five birds with similar mean bodyweight and bodyweight ranges. Twelve replicates were randomly allocated to each of four temperature-controlled rooms maintained at 31° ± 1°C. Three replicates in each room were randomly allocated to each of four treatments. In Experiment 1 all birds were fed the experimental diet containing 10.9 g lysine/kg with an arginine:lysine ratio of 1.21 (Table 1). The treatments were applied via the drinking water as follows:

- i) Control. Municipal town water
- ii) Town water containing 4.4 g L-arginine free base/L
- iii) Town water containing 5.6 g NaHCO₃/L
- iv) Town water containing 4.4 g L-arginine free base and 5.6 g NaHCO₃/L.

¹ Department of Animal Science, University of Sydney, Camden, NSW 2570.

² Department of Poultry Science, North Carolina State University, Raleigh, North Carolina 27695-7608, USA.

³ Nutri-Quest Inc., Chesterfield, Missouri 63017, USA.

In Experiment 2 all birds were fed the experimental diet containing 9.6 g lysine/kg with an arginine:lysine ratio of 1.22 (Table 1). The treatments were applied via the drinking water as follows:

- i) Control. Municipal town water
- ii) Town water containing 1.0 g L-arginine acetate/L
- iii) Town water containing 2.0 g L-arginine acetate/L
- iv) Town water containing 3.0 g L-arginine acetate/L.

Food and drinking water were supplied *ad libitum* to all birds for 21 days during which time production parameters were determined. Feed intake, liveweight gain and feed conversion measures were based on 12 replicates. Water consumption was determined on a room basis as the average of the three replicates on each treatment in each room and this measure was, therefore, based on 4 replicate values.

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

Ingredients	Experiment 1	Experiment 2
Sorghum	357.0	357.6
Wheat	451.8	451.8
Soyabean oil	3.3	3.3
Blood meal	20.0	20.0
Fish meal	47.1	47.1
Meat & bone meal	26.1	26.1
Soyabean meal	75.0	75.0
Ground limestone	10.2	10.2
Sodium chloride	1.2	1.2
Broiler premix	5.0	5.0
L-lysine HCl	0.9	0.9
DL-methionine	0.4	0.4
L-arginine acetate	-	1.4
L-arginine free base	2.0	-
<i>Determined analysis</i>		
Crude protein	208	195
Lysine	10.9	9.6
Arginine	13.2	11.7
Methionine	4.3	4.1
TSAA	7.9	7.6

Data were analysed by analysis of variance using the General Linear Model procedure (SAS). All variables were analysed using a completely randomised block design with the exception of the water intake and the water to feed intake ratio which were analysed using a completely randomised design. Where the treatment by block interactions was not significant the data for blocks were pooled. All statements of significance were based on a probability level of 0.05 and, where significant treatment differences were observed, the treatment means were partitioned using Duncan's multiple range test (SAS, 1988).

III. RESULTS

The results for Experiment 1 are shown in Table 2. Significant treatment effects were observed for all the measured parameters.

Table 2. Production and water intake responses of heat - stressed broilers (31°C) to sodium bicarbonate (NaHCO₃) and/or arginine free base (Arg) in the drinking water between 22 and 43 days of age in Experiment 1.

Water Supplement	Body weight gain (g)	Feed consumption (g)	Feed conversion (g:g)	Water intake ¹ (mL)	Water:Feed ¹ (mL:g)
None	1147 ^a	2241 ^a	1.955 ^a	5902 ^b	2.634 ^b
Arg	1046 ^b	2045 ^b	1.955 ^a	4747 ^c	2.326 ^c
NaHCO ₃	1156 ^a	2203 ^a	1.908 ^b	7155 ^a	3.253 ^a
Arg + NaHCO ₃	1020 ^b	2020 ^b	1.982 ^a	5648 ^b	2.789 ^b
SEM	19	37	0.015	236	0.112
<i>Probability</i>					
Room	NS	NS	NS	-	-
Treatment	0.0001	0.0001	0.008	0.0001	0.0001
Interaction	NS	NS	NS	-	-

^{a-c} Mean values within columns with different superscripts differ significantly ($P < 0.05$).

¹ Water intake determined as the mean of 3 replicates in each of 4 rooms.

The feed intake and liveweight gain of both arginine treatments were significantly reduced, and no improvement in feed conversion was observed, relative to the controls. Feed conversion was significantly improved by the NaHCO₃ supplement but this response was not observed with the combined NaHCO₃-arginine treatment. The water intake and the water:feed intake ratio of broilers receiving the arginine free base supplement were significantly reduced compared with controls. The water and the water:feed intake ratio were significantly increased in broilers receiving the NaHCO₃ but this response was not observed with the combined NaHCO₃-arginine treatment. The water intake and the water:feed intake ratio of the broilers receiving the combined arginine free base-NaHCO₃ supplement were similar to controls and significantly improved compared to broilers receiving the arginine supplement alone, but this was not reflected in improved production.

The results for Experiment 2 are shown in Table 3. There were no significant effects of the arginine acetate treatments on feed intake, bodyweight gain and feed conversion. However, there was a tendency for bodyweight gain and feed conversion to be adversely affected by the arginine acetate supplements. Water intakes of the birds receiving the 1.0 and 2.0 g/L arginine acetate supplements in the drinking water were reduced compared to controls, with the total water intake of the birds receiving the 2.0 g/L supplement being significantly affected. Water intakes of the control and 3.0 g/L supplemented birds were similar, as were weight gain and feed intake, but the feed conversion of the latter birds failed to respond positively to this increase in water consumption.

Table 3. Production and water intake responses of heat - stressed broilers (31°C) to arginine acetate (Arg Ac) in the drinking water between 21 and 42 days of age in Experiment 2.

Water Supplement	Body weight gain (g)	Feed consumption (g)	Feed conversion (g:g)	Water intake ¹ (mL)	Water:Feed ¹ (mL:g)
None	1168 ^a	2375 ^a	2.036 ^a	6384 ^a	2.685 ^a
Arg Ac, 1.0 g/L	1111 ^a	2310 ^a	2.086 ^a	6071 ^{ab}	2.629 ^a
Arg Ac, 2.0 g/L	1127 ^a	2342 ^a	2.080 ^a	5897 ^b	2.525 ^a
Arg Ac, 3.0 g/L	1144 ^a	2379 ^a	2.077 ^a	6356 ^a	2.686 ^a
SEM	20	28	0.085	140	0.064
<u>Probability</u>					
Room	0.023	0.001	NS	-	-
Treatment	NS	NS	NS	0.050	NS
Interaction	NS	0.013	NS	-	-

^{ab} Mean values within columns with no common superscript differ significantly ($P < 0.05$).

¹ Water intake determined as the mean of 3 replicates in each of 4 rooms.

IV. DISCUSSION

In contrast to the improvement in feed conversion observed when the arginine:lysine intake ratio was increased by supplementing the diet with arginine free base (Kroon *et al.*, 1997), the present results indicate that similar supplementation via the drinking water was unsuccessful. One explanation for the poor response to the arginine supplement may be the high pH of the drinking water solutions containing arginine free base which may have impacted adversely on feed consumption by reducing the intake of drinking water. Measurements of the drinking water used in each treatment gave the following results (Mean \pm SEM) for pH: (i) Control, 8.25 ± 0.53 ; (ii) Arginine free base, 9.94 ± 0.27 ; (iii) NaHCO_3 , 8.69 ± 0.12 ; (iv) Arginine free base + NaHCO_3 , 9.42 ± 0.05 .

In addition, the improvement in feed conversion observed with NaHCO_3 was negated by the simultaneous administration of arginine free base in the drinking water. The fact that the water intakes of broilers receiving the combined supplement was similar to controls and significantly improved compared to broilers receiving only the arginine supplement, whereas feed intakes were similar to those of broilers receiving only the arginine supplement and significantly reduced compared to controls, suggests that the poor feed consumption observed with the arginine free base treatments was not directly related to reduced water intake. In the second experiment the pH of the drinking water solutions were more normal and ranged between 7.75 and 8.09 and broiler performance was not improved by the addition of arginine acetate to the drinking water.

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PRODUCTION AND ECONOMICS OF FOUR IMPORTED AND TWO AUSTRALIAN LAYER STRAINS IN TWO CAGING SYSTEMS

J.V. NOLAN, J. R. ROBERTS, E. THOMSON, W. BALL and R.B. CUMMING

Summary

Egg production, egg quality and profitability of 6 strains of commercially available layers (four imported and two Australian strains) were evaluated at the University of New England's poultry farm 'Laureldale' in 1996-97. Mortality from Marek's Disease and cannibalism was much higher in imported birds (23-44%) than in Australian strains (5.7-7.7%). In this comparison, profitability differed markedly between strains: an Australian strain was the most profitable under our conditions when eggs were marketed by the dozen. Birds housed in one shed at 3/cage were more profitable than those in another shed at 5/cage.

I. INTRODUCTION

Imported strains of laying hens are likely to be subject to a wider range of environmental, disease and management stresses in Australia than in their countries of origin. High mortality of imported layers from Marek's disease and cannibalism have been major sources of economic loss in recent years.

A comparison was made at the University of New England in 1996-97 of the production and profitability of imported (Lohmann Brown, Hy-Line Brown, IsaBrown, HiSex) and Australian (Hy-Line CB, Tegel Black) strains of layers up to 66 weeks of age under management conditions that are typical of many Australian poultry farms.

II. MATERIALS AND METHODS

All birds were hatched between 16 and 30 January, 1996 and then subjected to identical treatment, after leaving their hatcheries of origin, until 66 weeks of age. All chickens were vaccinated at their hatcheries against Marek's Disease and Infectious Bronchitis (IB) and reared in wire-floored cages near Tamworth. At 3 weeks of age, they were re-vaccinated for IB (A3 virus; in-contact method) and at 14 weeks for Avian Encephalomyelitis and IB (Vic S; in contact). The birds were beak-trimmed at 10 days and at 8 weeks of age. At 17-18 weeks of age, they were moved to the 'Laureldale' poultry farm (University of New England) where they were housed in single-deck laying cages, at either 5 birds/cage (Harrison cages; Shed 1) or 3 birds/cage (Californian cages; Shed 2). At this stage, the birds were accustomed to 14 h daylight. Daylength was then increased in steps of 20 min/week to 16 h (at 24 weeks). Imported strains were selectively beak-trimmed at 31 weeks of age.

All strains were given, *ad libitum*, a commercial chick starter diet from 0 to 6 weeks, a commercial grower diet from 6 to 18 weeks and a commercial pre-layer diet from 18 to 22 weeks. From 22 weeks of age, all birds were offered a crumbled diet formulated to provide (per kg) 11.6 MJ of metabolizable energy, 175 g crude protein and 37 g calcium (Millmaster, Tamworth).

Department of Animal Science, University of New England, Armidale, NSW 2351.

Shed 1 had 11 replicates each of 40 birds for each of 4 strains, i.e. Hy-Line CB, IsaBrown, Tegel Black and Hisex strains were each represented by 440 birds (5 birds/cage in 88 cages), whereas the Lohmann Brown and Hy-Line Brown strains were represented by 12 replicates and 480 birds/strain. In Shed 2, each strain had 4 replicates each of 33 birds (132 birds/strain). The replicates were evenly placed throughout each shed.

Birds that died were replaced up to 25 weeks of age. Post-mortem examination was made on all birds that died to 66 weeks of age. Feed intakes and egg productions of birds were determined from 22 to 66 weeks of age. Eggs were collected (30/strain) at random from all treatments at intervals of 4 weeks for egg weight measurements. Costs and returns were recorded to enable net returns per bird housed to be determined.

Results were analysed statistically by analysis of variance.

III. RESULTS

Mean liveweight of pullets at 18 weeks of age ranged from 1.48-1.75 kg, increasing to 2.18-2.37 kg at 66 weeks of age.

Cumulative mortalities for each strain across treatments and sheds are given in Fig. 1.

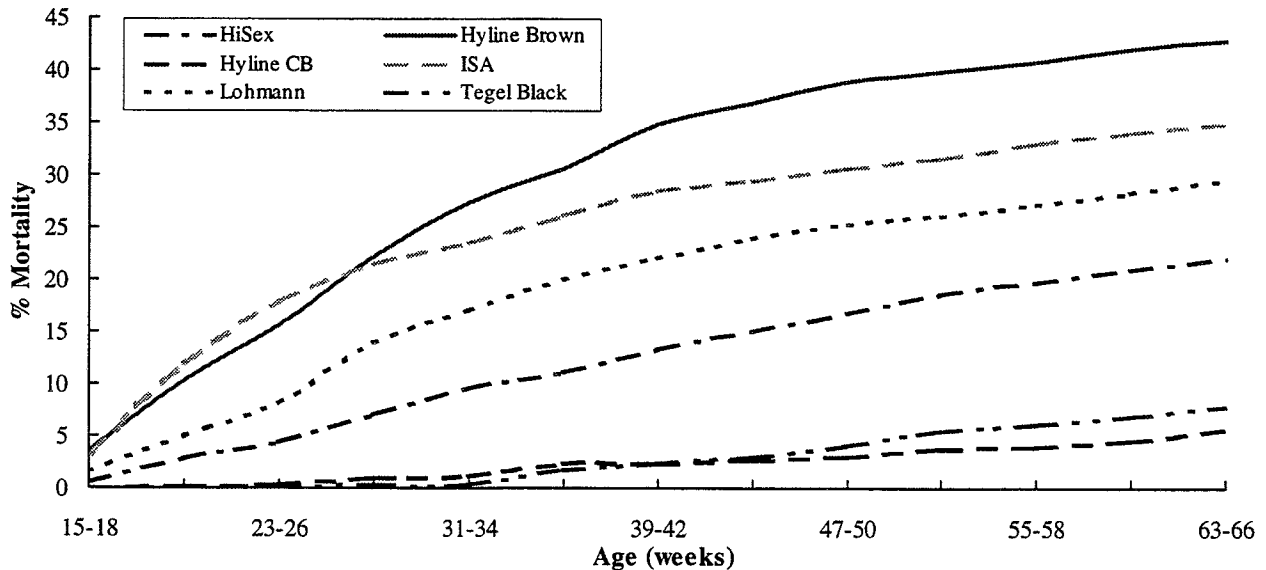


Figure 1. Cumulative mortality in each of 6 strains of layers from 18-66 weeks of age.

Post-mortem examinations indicated that the majority of deaths were due to Marek's disease (MD) and cannibalism. Of the imported strains, the Hy-Line Brown and IsaBrown had the highest incidence of MD, whereas the Hisex and Lohmann birds showed some MD resistance: deaths due to cannibalism occurred in all imported strains and were greater ($P < 0.05$) in 5-bird than 3-bird cages, and highest for Lohmann birds. The Australian genotypes were almost totally free of MD and the 2-3% of deaths attributed to cannibalism was apparently not affected by the number of birds/cage. Lohmann Brown, HiSex and Hy-Line Brown hens were first into lay, reaching 50% hen-day production at 21-22 weeks of age. All strains reached peak hen-day production at about 27 weeks of age. Hen-day egg production peaked at over 90% for the Australian strains, and between 84 and 90% for the imported strains. Hen-day production declined more rapidly in the Australian strains and was lower ($P < 0.05$) than in the imported strains after 45 weeks of age. Differences ($P < 0.05$) in

hen-housed egg production between the strains were smaller than in hen-day egg production as the fall in egg production from 45-66 weeks in the Australian strains was offset by their lower mortality (Figure 2). Egg weight increased in all strains as the hens aged, but the Australian strains laid lighter eggs at all times (Figure 3) and, as a consequence, had a lower egg mass production at all times (Figure 4). Feed intake, and feed conversion ratio (feed intake/eggmass) were higher ($P < 0.01$) in the two Australian genotypes than in the imported strains (Table 1).

Table 1. Cumulative mortality (% hens housed), hen-day and hen-housed egg production, mean egg weight, feed intake, total eggmass production (kg), and feed conversion ratio (FCR, g feed/g eggmass) in 6 strains of layers (from 22 to 66 weeks of age).

	Hisex	Hy-Line Brown	IsaBrown	Lohmann	Hy-Line CB	Tegel Black
Mortality (%)	21.4	37.5	30.2	27.8	5.6	7.9
Eggs/hen-day (%)	81.5	84.9	78.7	82.5	77.6	77.1
Eggs/hen housed	222	188	200	191	237	231
Egg weight (g)	63.6	63.2	63.2	64.0	56.3	59.2
Eggmass prod. (kg)	14.5	13.6	13.8	14.2	13.7	13.1
Feed intake (g/d)	133	137	129	132	134	135
FCR (g/g)	2.58	2.54	2.61	2.50	3.11	3.00

Profitability was assessed by subtracting costs (of rearing pullets, including replacements between 18 and 25 weeks, feed and packaging materials, but excluding overheads, labour and other running costs) from income (from egg sales and spent birds) (Table 2).

Table 2. Net returns (\$ per hen housed) in Shed 1 (5-bird cages) and Shed 2 (3-bird cages) for 6 strains of layers (income and costs from hatching to 66 weeks of age).

	Hisex	Hy-Line Brown	IsaBrown	Lohmann	Hy-Line CB	Tegel Black
Shed 1	3.54 ^φ (4.63)*	2.03 (5.16)	1.57 (5.31)	3.47 (5.68)	4.17 (1.63)	2.90 (1.75)
Shed 2	3.92 (6.07)	2.68 (5.88)	2.13 (5.34)	5.14 (7.32)	5.93 (3.85)	4.46 (3.80)

Net return = [return from egg sales + spent hens] - [costs of rearing + feed + egg packaging]

^φAssuming all eggs sold by the dozen @ \$1.30. *Assuming all eggs sold by the kg @ \$1.725/kg.

IV. DISCUSSION

The high mortalities among imported strains confirm the Australian industry experience and our earlier research (Nolan *et al.*, 1997) which show there is still a major mortality problem with the recently imported strains. The imported strains are, however, capable of feed-efficient egg production which will be further improved if the mortality problems can be overcome. The profit margin obtained for each strain depended on the method used to market eggs. Selling 'by the dozen' in Armidale was more profitable for the Australian strains despite the higher packaging costs, whereas selling 'by the kg' was more profitable for imported strains. The relative returns from egg sales can markedly affect the profitability ranking of the birds.

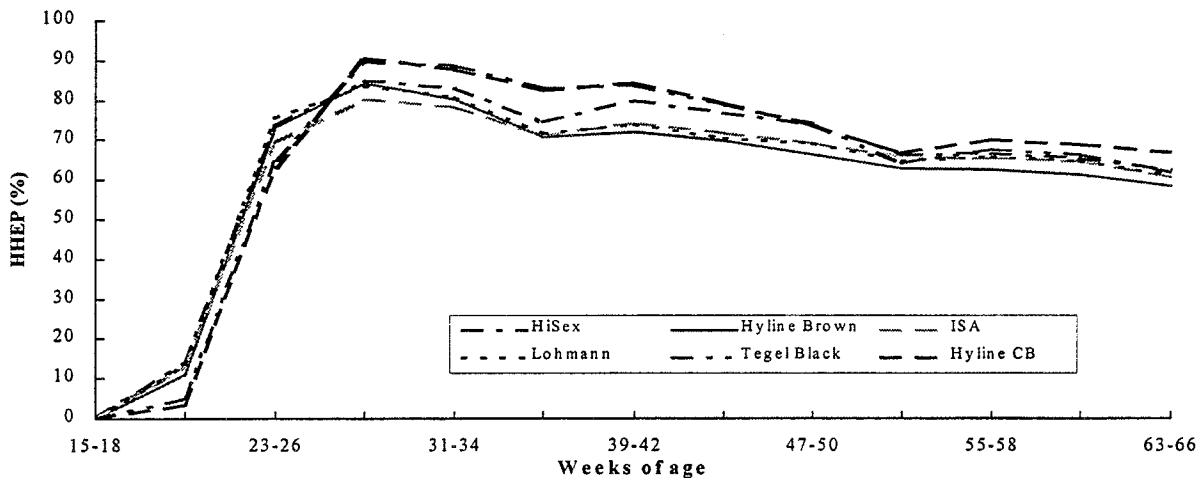


Figure 2. Differences in hen-housed egg production (HHEP) in 6 strains of layers to 66 weeks

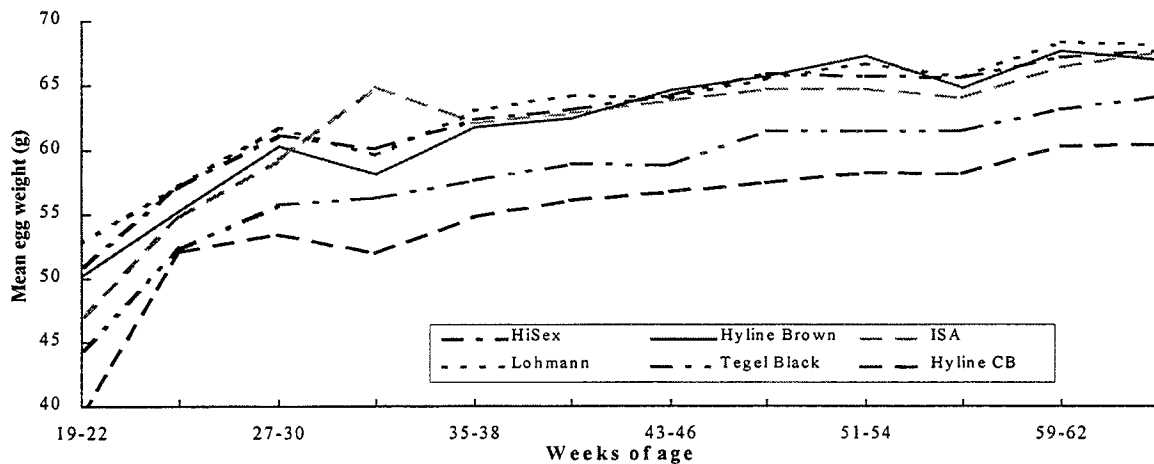


Figure 3. Differences in mean egg weight with increasing age between strains of layers

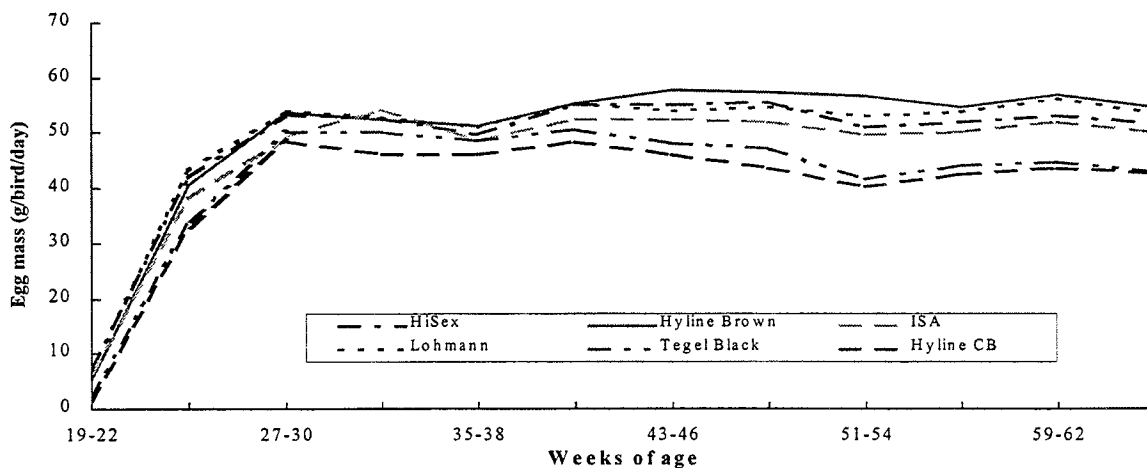


Figure 4. Eggmass production of 6 strains of layers from 22-26 weeks of age

Continued assessment of mortality, especially as related to the effects of the number of birds per cage, and feed conversion efficiency, will be essential. Crossing of the MD-resistant Australian strains with imported genotypes, development of more efficient vaccines, or raising young birds in isolation may be ways of reducing economic losses due to high MD-related mortality. Ways of reducing cannibalism also need to be improved. Further studies of this type will be needed to provide experimentally controlled, early and independent evaluations of new strains of layers under Australian conditions.

V. ACKNOWLEDGEMENTS

We thank the Egg Industry Research and Development Council for financial assistance and the companies (Barter, Biaida, Hy-Line and Tegel) and the NSW Egg Producers Co-Operative for their collaboration, advice and 'in-kind' support.

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THE EFFECT OF HOUSING ON THE BEHAVIOUR OF CHICKENS

C. RUDKIN and J.G. DINGLE

Summary

The time spent in various activities by ranging growing pullets and hens of a commercial strain in semi-natural conditions was measured. These data were then compared with reported activities of Junglefowl ranging in semi-natural conditions, and of hens in various housing conditions.

It was found that the time spent foraging by the commercial strain was about the same as that by Junglefowl. Foraging rates were reduced, and feeding rates increased in more intensive housing systems.

Birds were shown to modify their behaviour in intensive husbandry systems, but the reported increase in feather pecking under these conditions suggests that increased pecking at food does not compensate the birds for loss of the opportunity to forage.

I. INTRODUCTION

An important but rarely considered need is to understand the behaviour of animals in environments to which they are adapted by natural selection, in order to obtain information about what conditions are likely to result in good welfare (Dawkins, 1989; Broom and Johnson, 1993, p 146). It was therefore decided to observe the time spent in various activities (time budgets) of growing pullets and adult hens of an Australian commercial breed in semi-natural conditions. These were then compared with time budgets of Junglefowl and of commercial breeds in various housing conditions reported in the literature.

II. METHODS

Nine SIRO-CB day-old chicks with untrimmed beaks were housed in an enclosed pen with food and water provided *ad libitum*.

The chickens were allowed to range freely outside the pen during the day from the fourth week. The environment contained areas sheltered by various trees (a large Jacaranda, Moreton-Bay Fig, Bougainvillea, Mulberry, and three Queensland Nut trees). Substrate consisted of areas of bare sandy loam, leaf-litter, tall weeds, and couch-grass.

Chickens were observed from the fifth week after hatching to the 18th week (pullets), and from the 19th to the 26th week (hens). They were observed for three two-hour sessions a day, and two days each week. Observations consisted of five-minute scans. Thus there were 24 scans in each session, and 144 scans each week. At each scan, as each chicken was located and identified, it was observed for a few moments, and its prime activity noted.

Behaviours were recorded under the following categories:

Environmental pecking: standing or moving about, pecking at a variety of objects.

Grazing: most pecks at blades of grass which were plucked and eaten.

Scratching and pecking: scratching the ground and pecking at uncovered material.

These first three categories are the components of foraging.

Food pecking: pecking at the food provided in the pen.

Drinking: drinking water from any source.

Sitting: sitting with keel on substrate.

Standing: quietly standing.

Standing and preening: quietly standing and preening.

Dust-bathing: sitting in dust-bath.

Nesting: from when the bird left the flock to find a nest, to when she left the nest.

Other: behaviours that did not fit readily into any of the above categories.

The mean proportion of scans when each behaviour was recorded for each week and, as pullets and as hens, was calculated. The mean weekly number of consecutive scans (run length) of each behaviour was also calculated.

III. RESULTS

Time spent sitting was less at ages six to eight weeks, 11 to 12 weeks, and dropped markedly towards onset of lay until, from the 20th week, only a very small proportion of the time was spent sitting (Figure 1). Time spent dust-bathing, as well as run length of dust-bathing, tended to increase at ages when there was a reduction in sitting time.

Proportion of time feeding, as well as run lengths tended to be less at six to eight weeks as well as at 18 to 21 weeks (Figure 1), and run length of feeding behaviour also lessened at 10 to 13 weeks. On the other hand, the various foraging-type behaviours tended to increase at these times (Figure 1). The run length of environmental pecking increased at 18 weeks, proportion of time spent grazing increased at 11 to 12 weeks, and proportion of time spent scratching and pecking increased from the 18th week.

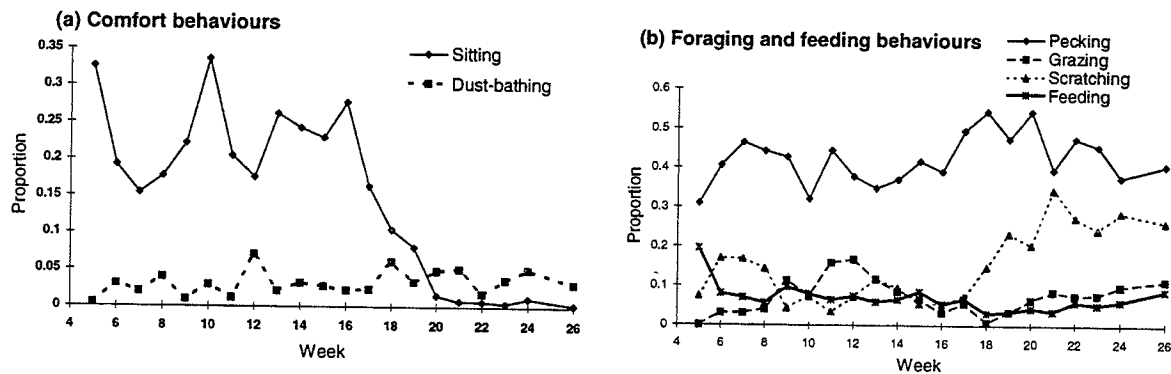


Figure 1. (a) Mean weekly proportion of scans in which comfort behaviours were recorded; and (b) mean weekly proportion of scans in which foraging behaviours and feeding were recorded.

Figure 2 shows the time budgets seen in this study of pullets and of laying hens, and compares these with time budgets of birds reported in other studies. Our birds foraged as much as Dawkins' (1989) Junglefowl. Litter housed hens walked and foraged less than ranging hens, and sat and fed more. Birds on slats foraged less and fed more than those on litter, and birds in cages stood more than those on slats.

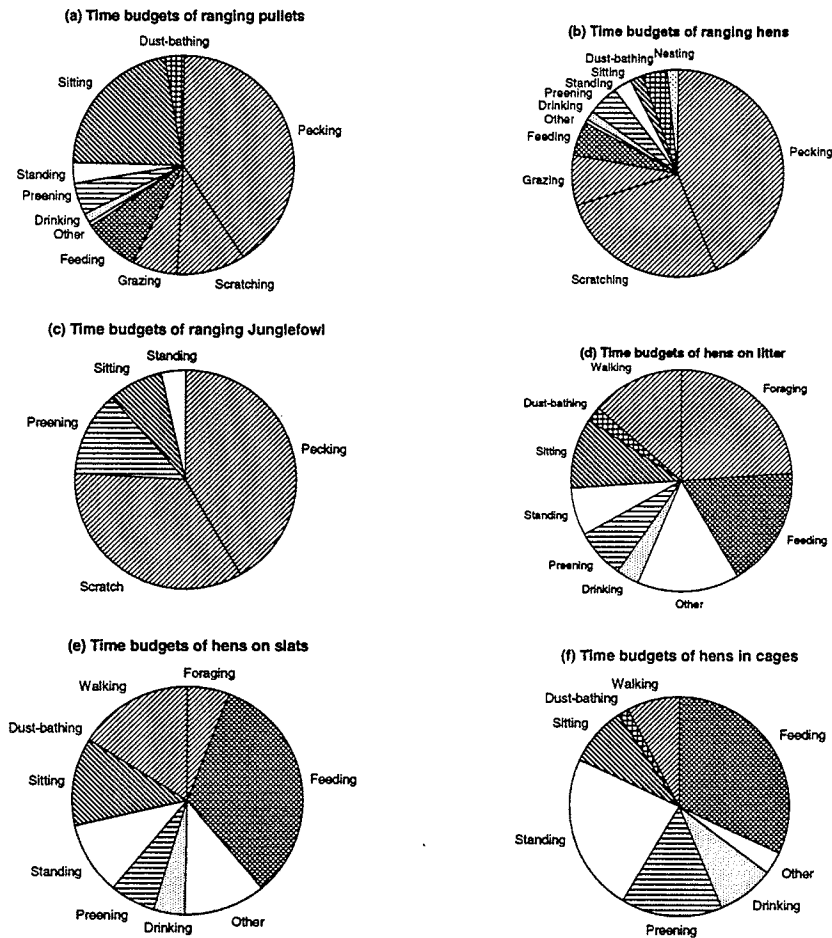


Figure 2. Time budgets of: (a) pullets to 18 weeks; (b) hens from 19 to 26 weeks; (c) Junglefowl in semi-natural conditions (data from Dawkins, 1989); (d) hens on litter (data from Appleby *et al.*, 1989; (e) hens on slats (data from Appleby *et al.*, 1989); (f) hens in cages (data from Braastad, 1990).

IV. DISCUSSION

These results have major implications for housing. This domestic strain appears little changed from the ancestral Junglefowl in the amount of foraging it performs. Lower foraging rates and increased sitting and feeding rates on litter, compared to rates in semi-natural conditions, indicate that litter housing modifies the natural behaviours of the birds. Behaviour rates on non-litter floor housing were more similar to those in cages than on litter. The comparatively low sitting rate in cages (less than ranging pullets) indicates that hens do not adapt to cages by filling their extra time by sitting. It is paradoxical that the age at which the birds become very much more active with scratching and foraging under 'natural' environments, is the age that they are transferred to cages.

Time-budgets of the ranging hens also have implications for current attempts to improve housing for hens. Enriched cages provide dust-baths and nests for the birds, yet these only occupy a small proportion of the day of ranging hens.

Nicol and Dawkins (1990) note that most "free-range" farms provide open grassland for birds rather than dense varied vegetation, and that birds generally remain close to the house. Hens in the present study grazed for only 7.9% of the time, and it was noticed that the birds only ventured out onto grass that was under the shade of trees, or onto open grassy areas

when the sun was behind clouds or low on the horizon. Thus even free-range farms may not provide environments which facilitate the performance of 'natural' behaviours.

The ages at which sitting time is reduced, and foraging is increased, are the ages at which the risk of feather pecking increases (Hughes and Duncan, 1972). However, feeding rates did not increase at these ages. Thus feather pecking may be more to do with a foraging need than a feeding need. Other results also indicate that feather pecking is associated with foraging behaviour (eg Blokhuis and van der Haar, 1992). Other factors that influence feather peck rates do so by acting on the motivation or opportunity to forage. The increased food pecking by more intensively housed birds represents foraging activity rather than feeding activity (Appleby *et al.*, 1992). The observation that feather pecking is worse under barren conditions (Appleby *et al.*, 1992) indicates that food pecking is not a satisfactory substitute for 'natural' foraging activity.

V. CONCLUSIONS

All modern commercial housing systems are very well designed to provide the maximum possible comfort and safety for the birds. However, they are not designed to allow birds to perform 'natural' behaviours, and the evidence is that the birds significantly alter their behaviours in different housing conditions.

It is suggested that attempts to improve housing for birds should take into account their time budgets in semi-natural conditions. Foraging occupies a large proportion of the day in these birds. Dawkins (1990) notes that the demand for the opportunity to peck and scratch at litter is "inelastic" and that hens will "pay a price" to be able to carry out the 'natural' feeding behaviour. Therefore, housing that provides an opportunity to express foraging behaviour may improve conditions for the birds.

Rudkin (1997) showed that it is possible to enrich present housing systems relatively simply by including racks with lucerne hay. It was suggested that exploratory foraging is the important component not met by the extra food pecking in the more intensive systems. Lucerne hay gave the birds something to peck and investigate to see if it was edible. It reduced feather pecking and almost eliminated cannibalism by pullets in pens, and completely eliminated cannibalism by hens in cages.

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THE EFFECT OF LINOLEIC ACID IN LAYING HENS LATE IN LAY

A.M. LEARY, J.R. ROBERTS and W. BALL

Summary

Six strains of laying hen, including four imported and two Australian strains, were fed diets containing varying concentrations of linoleic acid from nine weeks of age. Dietary levels of linoleic acid included a low, control and high level of linoleic acid which were 6, 13 and 30g/kg, respectively. Early in lay the effect of linoleic acid on egg weight was strain dependent, with the Hy-Line CB Black, Tegel SB2 and Hi-Sex Brown hens responding to varying dietary concentrations of linoleic acid. Later in lay, from 44 to 60 weeks of age, the Hy-Line Brown, Lohmann Brown and Hy-Line CB Black hens also produced significant responses to changes in dietary linoleic acid. These results indicate that the response of laying hens to varying dietary levels of linoleic acid late in lay is strain dependent.

I. INTRODUCTION

Recently Australia has begun importing strains of laying hens that lay brown eggs, which the Australian consumer prefers. However one of the problems associated with these birds is the large size of the eggs they produce. This is particularly a problem as egg size increases as the birds age, (Cowen *et al.*, 1964) and eggs produced late in lay are much too large for the Australian market (Robinson, 1996).

Two main areas have been investigated to try to reduce egg size in the imported strains of laying hens, namely age of light stimulation and diet. Early light stimulation has been shown to reduce egg size throughout the entire laying life of the bird (Morris, 1994). It appears that this occurs as a result of a low body weight as the birds begin lay. However there is anecdotal evidence that a low body weight at point of lay can make the birds more susceptible to Marek's disease.

The nutrient content of the diet is very important in manipulation of egg size. Diets high in both fat and protein have been shown to increase egg size (Kerhavarz, 1995). Another study has indicated that by increasing protein in the diet a proportional increase in egg size may occur (Morris and Gous, 1988). Also a reduction in the amount of methionine in the diet has been correlated with a decrease in egg size (Petersen *et al.*, 1983).

Egg size is affected by dietary linoleic acid, which is an essential fatty acid in chickens. The effect of linoleic acid on egg size was discovered when it was found that an unknown factor in diets high in corn oil or rice pollard increased egg size (Jensen *et al.*, 1958). This unknown factor was later shown to be linoleic acid (Balnave, 1972; Scragg *et al.*, 1987; Mannion *et al.*, 1992).

The present study has investigated whether reductions in dietary linoleic acid produce a resultant reduction in egg size in imported and Australian strains of laying hen, especially late in lay when egg size in the imported strains exceeds industry requirements. It is hypothesized that if an increase in dietary linoleic acid increases egg size, then a decrease in linoleic acid may have the opposite effect.

Division of Animal Physiology, School of Rural Science and Natural Resources, University of New England, Armidale, 2351.

II. METHODS

The current experiment consisted of 18 groups of hens (approximately 18 birds/group) made up of six strains and three dietary treatments. The strains involved in the experiment were four imported strains (Isa Brown; Lohmann Brown; Hi-Sex Brown and Hy-Line Brown) and two Australian strains (Tegel Super Brown and Hy-Line CB Black). The birds were reared on a broiler starter diet and at nine weeks of age were randomly divided into three groups and given diets containing either 6, 13 or 30g/kg linoleic acid. Diets were formulated to have the same metabolisable energy and nutrient constraints. Eggs were collected periodically throughout lay at 14-20, 23, 26, 31, 36, 44, 52 and 60 weeks of age.

The following measurements were made:

Egg Weight	Length	Width	Shell Colour
Shell Reflectivity	Breaking Strength	Deformation	Haugh Units
Yolk Colour	Yolk Weight	Shell Weight	Shell Thickness

Only the results for egg weight are presented in this paper.

III. RESULTS

Table 1. The effect of dietary linoleic acid on egg weight in six strains of laying hen.

Strain	Diet* g/kg	Weeks of Age							
		14-20	23	26	31	36	44	52	60
Isa Brown	6	45.1	55.3	57.9	60.7	61.6	63.6	65.3	65.0
	13	43.7	56.9	59.0	61.4	63.8	65.1	66.9	65.6
	30	43.9	54.9	58.9	60.6	61.1	64.4	66.1	67.5
Loh- mann Brown	6	48.0	57.4	60.9	64.1	65.9	65.5	69.0	68.3 ^a
	13	47.3	56.7	59.1	62.8	64.6	67.0	68.9	67.0 ^a
	30	45.9	58.6	61.3	64.1	66.6	69.7	71.1	72.1 ^b
Hy- Line Brown	6	46.2	54.1	57.6	60.5	61.6 ^b	63.2 ^b	65.5 ^b	67.0
	13	48.1	56.4	59.2	63.4	64.8 ^a	68.2 ^a	69.9 ^a	70.0
	30	46.5	55.9	60.1	62.9	63.4 ^{ab}	67.2 ^a	68.0 ^{ab}	68.2
Hi-Sex Brown	6	50.1	56.4 ^b	60.4	64.0	66.1 ^{ab}	67.8	68.7	68.6
	13	49.3	56.7 ^b	60.4	63.0	63.5 ^b	67.4	68.3	69.5
	30	50.0	61.5 ^a	62.1	65.7	67.9 ^a	70.7	71.2	72.2
Hy-Line CB Black	6	40.8	48.5	50.8 ^b	53.2 ^b	55.7 ^c	57.7 ^c	60.5 ^b	60.7
	13	40.8	48.5	52.2 ^b	55.0 ^a	57.1 ^b	59.9 ^b	62.5 ^a	62.9
	30	42.4	50.5	54.1 ^a	56.2 ^a	58.5 ^a	61.7 ^a	62.8 ^a	63.6
Tegel SB2	6	45.2	50.0 ^b	55.0 ^b	58.4 ^b	60.3	61.8	62.9	64.5
	13	45.2	57.2 ^a	57.8 ^{ab}	60.4 ^b	63.2	63.3	66.5	68.3
	30	46.7	54.8 ^a	60.0 ^a	64.2 ^a	64.1	64.1	63.3	64.1

* Linoleic acid content of the diet (g/kg)

Values within a strain and age at sampling with different superscripts are significantly different from one another ($P < 0.05$).

Egg weight in the Isa Brown hen was not significantly affected by the concentration of linoleic acid in the diet (Table 1). There was no significant difference in egg weight found between the different treatment groups in the Lohmann Brown hen until the final collection where the high linoleic acid diet produced the heaviest eggs. The Hy-Line Brown hens on the low linoleic acid diet produced significantly lighter eggs between 36 and 52 weeks of age. The Hi-Sex Brown birds on the high linoleic acid diet produced heavier eggs throughout the course of lay, although these results were only significant at 23 and 36 weeks of age. The Hy-Line CB Black hens produced significantly lighter eggs on the low fat diet between 26 and 52 weeks of age and significantly heavier eggs on the high linoleic acid diet at 36 and 44 weeks of age. The Tegel SB2 hens produced significant results in the early collections (23 to 31 weeks of age) with eggs produced on the high linoleic acid diet being significantly heavier than those produced by the hens on the low linoleic acid diet.

IV. DISCUSSION

The results do not support the hypothesis that low dietary linoleate levels reduce egg weight. They do indicate however that linoleic acid influences egg weight in some strains both early and late in lay. The weight of eggs produced by the Isa Brown hens however did not change in response to varying concentrations of dietary linoleic acid. The Lohmann Brown and the Hi-Sex Brown hens were affected by high levels of linoleic acid in the diet as egg weight increased. The Hi-Sex hens were more affected early in lay, whereas the Lohmann Brown birds began to respond significantly in the final collection at 60 weeks of age. The results for these three strains of imported birds indicate that linoleic acid is not effective in controlling egg weight either early or late in lay. The Hy-Line Brown birds were affected late in lay and reacted with a drop in egg weight as the amount of dietary linoleic acid fell. The most consistent reaction to changes in dietary linoleic acid was found in the Hy-Line CB Black hens which produced lighter eggs on the low linoleic acid between 26 and 52 weeks of age and significantly heavier eggs on the high linoleic acid diet at 36 and 44 weeks of age. Since the Hy-Line CB Black hens produce small eggs, the relatively consistent response in egg weight to increased dietary linoleic acid could be put to good use commercially with this strain. The Tegel SB2 hens responded to low levels of dietary linoleic acid with reduced egg size early in lay.

V. CONCLUSION

The effect of dietary linoleic acid on egg weight throughout lay is strain dependent with the Hy-Line CB Black hens being responsive both to increases and decreases in dietary linoleic acid. The only one of the imported strains in which egg weight reduction was achieved by reducing dietary linoleic acid is the Hy-Line Brown hen. Manipulation of dietary linoleic acid does not appear to be a generally effective method of reducing egg size in imported strains of laying hens late in lay.

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SEQUENTIAL STUDIES OF SKELETAL CALCIUM RESERVES AND STRUCTURAL BONE VOLUME IN A COMMERCIAL LAYER FLOCK

P.H. CRANSBERG¹, E.C. JONGMAN¹, B.H. THORP², S. WILSON³,
J. GOLDSMITH⁴, A. ALMOND⁴ and G.B. PARKINSON¹

Summary

Production induced osteoporosis in caged laying hens has been speculated to represent a major constraint to continuing genetic development. Models need to be developed that describe the induction of osteoporosis, and its interaction with environmental and management factors. The current paper monitors bodyweight, egg production, femur calcium and bone histology in a commercial flock of ISA Brown layers from 16 through to 68 weeks of age.

Flock bodyweight declines between 35 and 45 weeks of age, which was correlated with a loss of skeletal calcium reserves (15-20%) and with the induction of osteoporosis. As egg production decreased between 42 and 68 weeks of age, birds were able to replete skeletal calcium to levels achieved in early egg production. It seems likely that better standardisation of the equilibrium between growth, skeletal reserves, feed intake and egg production can reduce osteoporosis, as well as improving the productive potential of modern laying strains.

I. INTRODUCTION

Research has demonstrated a high incidence of bone fragility in caged laying hens. This has been mainly attributed to the development of osteoporosis (Riddell, 1981), which has been speculated to represent a serious constraint to further genetic development in the commercial laying hen. In general terms osteoporosis is defined as a local or systemic deficiency in the quantity of fully mineralised structural bone (Thorp, 1994), and has been described histologically as a reduction in the volume of structural trabecular and cortical bone within the skeleton. The vertebral skeleton, ribs and pelvis appear most susceptible to the development of osteoporosis, whilst the femur is the most susceptible part of the appendicular skeleton (Taylor and Moore, 1954; Riddell, 1981). Pathology characteristic of osteoporosis is clearly evident in the laying hen by about 40-45 weeks of age (Wilson *et al.*, 1992), which corresponds to the period of peak egg mass output in mid egg production.

The potential for production induced osteoporosis has been largely disregarded in more recent research, despite historical evidence that flock management can play a critical role in influencing skeletal mineralisation. Early studies on cage layer fatigue indicated some predisposing management factors, including precocious egg production and egg production in under weight pullets in warm weather (Grumbles, 1959). The normal metabolic equilibrium in the fowl can therefore be disrupted by mismanagement, and disproportionately high levels of egg production can occur in birds which are underweight with low tissue reserves and low nutrient intakes. Furthermore, there is likely to be an important role for calcium nutrition and calcium turnover in determining the mineral content of the skeleton, but the relationship of calcium turnover to the development of osteoporosis is poorly understood.

¹ Victorian Institute of Animal Science, Attwood, Victoria, Australia.

² Ross Breeders, Midlothian, Scotland, U.K.

³ Roslin Institute, Edinburgh, Midlothian, Scotland, U.K.

⁴ Victorian College of Agriculture and Horticulture, Longerenong, Victoria, Australia.

Maintenance of adequate skeletal calcium reserves is believed to be integral to sustaining production and shell quality, but there is little quantitative data on these parameters.

In an attempt to begin clarifying some of these relationships this paper reports on a flock of ISA Brown layers sequentially studied for growth, egg production, femur calcium reserves and bone histology.

II. METHODS

The ISA Brown flock was housed at commercial density (4 birds per cage) in a controlled environment shed where the temperature was maintained between 20-25°C. Birds were fed a commercial layer ration of 180 g/kg crude protein and 11.80 MJ/kg of metabolisable energy to 40 weeks of age, then 165 g/kg crude protein and 11.38 MJ/kg of metabolisable energy for the remainder of the experimental period. Calcium and phosphorous levels were included in the diet at 38 g/kg and 7 g/kg respectively.

Birds were monitored weekly for bodyweight up until 42 weeks of age, and then intermittently. Egg production was recorded weekly between 16 and 65 weeks of age.

Twenty left femur samples were obtained at 16, 23, 31, 42, 48 and 68 weeks of age and stored frozen until analysed for calcium content. The same twenty birds also had samples of the proximal tarsometatarsus removed for embedding and histological examination. The bone sections were examined blind and histomorphometry was used to quantify the percentages of medullary and trabecular bone (Wilson *et al.*, 1992). At each sampling age the birds were examined for nodulation and deformity of the rib cage, with the incidence of osteoporosis calculated as a percentage of the twenty bird sample based on the subjective assessment of the rib cages.

III. RESULTS

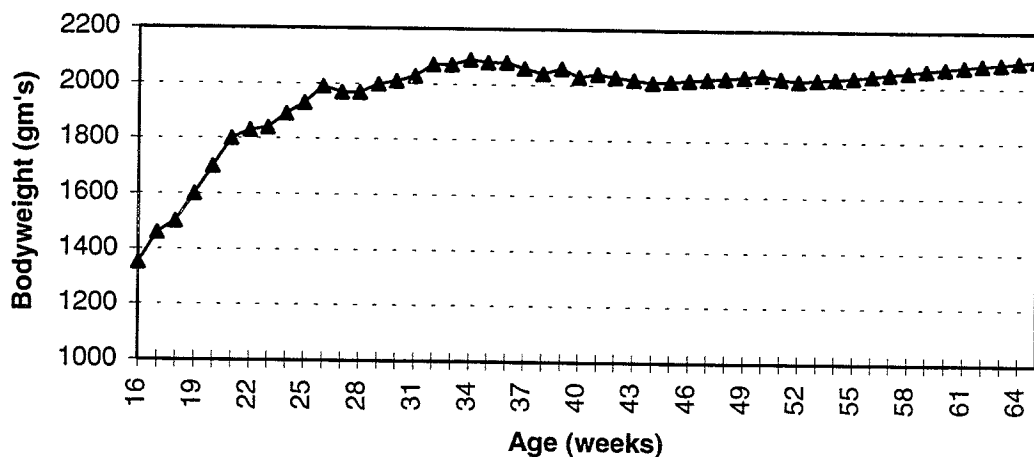


Figure 1 Average Bodyweight of ISA Brown Flock from 16-66 Weeks of Age
(Note: following 42 weeks of age birds were weighed at 44, 50, 52, 56 and 65 weeks of age. Weights between were estimated assuming linearity)

The sequential studies of flock growth illustrated a period of sub-optimal growth in early egg production (21-28 weeks) followed by a period of weight loss between 34-38 weeks of age (Figure 1). This growth pattern has been recorded in a large number of commercial

layer flocks in Victoria, and is most notable in flocks housed in controlled environment shedding. The peak flock weight was 2.1 kg at 35 weeks of age.

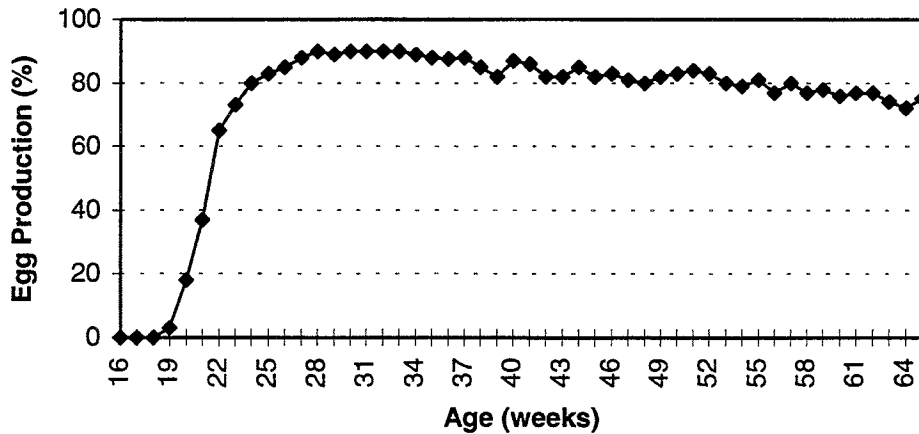


Figure 2. Egg Production of ISA Brown Flock.

Egg production peaked at 90% in weeks 28 and 30-33, but decreased in association with the decrease in bodyweight between 34 and 38 weeks of age (Figure 2).

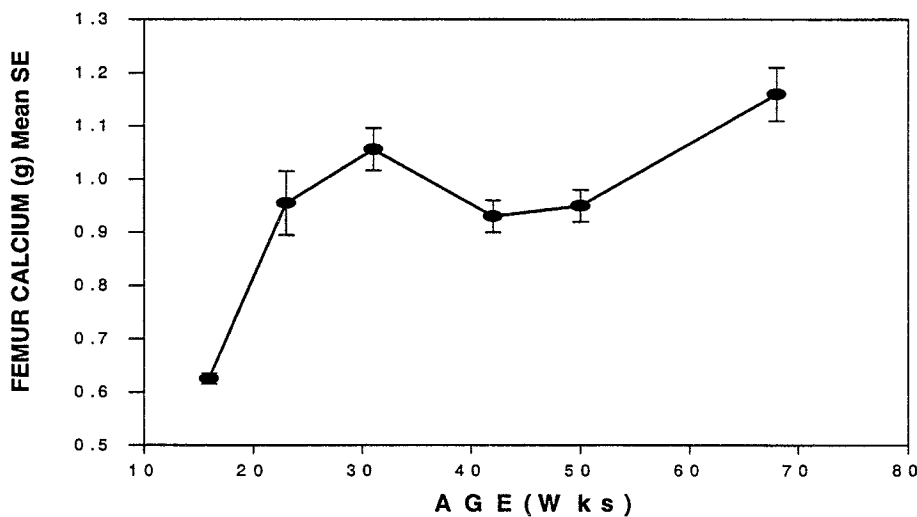


Figure 3 Average Femur Calcium Content (grams/bone) of ISA Brown Flock from 16-68 Weeks of Age (Mean (SE)).

Femur calcium content increased by over 50% from 16-23 weeks of age as the birds began egg production, and peaked at 31 weeks of age. The femur calcium then declined significantly ($P < 0.05$) by 42 weeks of age. The decline in femur calcium appeared correlated with the loss of bodyweight and the decline in egg production occurring in mid-lay. The loss of bodyweight (5%) and femur calcium (20%) corresponded to the theoretical peak egg mass output (35-45 weeks of age). From 48 to 70 weeks there is a rise in femur calcium associated with the decrease in egg production.

The comparison between femur calcium content and bone density parameters (trabecular bone volume and medullary bone volume) indicates that the shift in calcium

balance is well correlated with the change in both structural and mobilisable bone volumes of the proximal tarsometatarsus (Table 1). In the period from 31-42 weeks of age trabecular bone volume decreased by 50% and medullary bone decreased by 75%, followed by an increase to 48 weeks of age (Table 1).

Table 1. Sequential Determination of Femur Calcium (grams/bone), Trabecular Bone Volume (TBV%) and Medullary Bone Volume (MBV%) in the Proximal Tarsometatarsus of the ISA Flock (\pm standard error).

Age (weeks)	Femur Calcium (gram's/bone) Mean (SE)	Trabecular Bone Volume (%) Mean (SE)	Medullary Bone Volume (%) Mean (SE)
16	0.65 \pm 0.01	21.90 \pm 1.40	2.38 \pm 0.28
23	0.95 \pm 0.04	15.06 \pm 0.92	2.94 \pm 0.42
31	1.05 \pm 0.06	11.37 \pm 0.91	7.24 \pm 0.59
42	0.93 \pm 0.03	5.24 \pm 0.58	1.73 \pm 0.19
48	0.95 \pm 0.03	12.21 \pm 0.77	4.66 \pm 1.64
68	1.16 \pm 0.05	*	*

The nodulation and deformation of the rib cage, characteristic of osteoporosis, was initially evident at 42 weeks of age with 80% of birds affected. At 68 weeks of age both the incidence of rib abnormalities and the severity of the rib nodulation and deformity remained unchanged.

IV. DISCUSSION

In the model presented in this research it is clear that there was a profound shift in skeletal calcium reserves in mid-lay (31-42 weeks of age). The onset of osteoporosis is likely to occur in this period when skeletal calcium reserves are at their lowest. Histological evidence indicates that trabecular bone volume had declined by approximately 50% in a similar time frame, suggesting that the birds had used structural bone to sustain production, thereby increasing their susceptibility to osteoporosis. Under the pressure of continuously high egg mass output, the flock lost approximately 80-100 grams live weight (5%) and approximately 15-20% of total femur calcium content. If the femur calcium loss is representative of the whole skeleton then approximately 50% of the mobilisable pool of calcium (4-5 grams calcium) had been depleted between 31-42 weeks of age. As the pressure of egg mass output declined, the skeleton was able to replenish calcium stores and both trabecular and medullary bone (Rennie *et al.*, 1997).

Clearly there are likely to be large shifts in skeletal mineral content occurring in mid-lay which are proportionally larger than the shifts in liveweight. The model developed is likely to be representative of commercial flocks and can form standards for additional analysis. Evidence collected in more recent studies indicates that the magnitude of the suppression of liveweight gain in early and mid-lay may not always be as severe as in these studies, but the age of the growth depression was very consistent across flocks.

Considering the magnitude of the shifts in skeletal mineralisation that could be occurring in mid-lay, there may be a relationship between the persistency of production, shell quality and calcium metabolism which could be moderated by new nutritional approaches. Furthermore, the close association between the development of the osteoporosis in mid-lay

and the drain of production, supports the premise that the production induced osteoporosis is much more significant in the induction of bone fragility in caged hens than the confinement and reduced mobility associated with the cage system. In future, comparisons of bone fragility or density should be carefully standardised for egg mass output.

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EFFECT OF CALCIUM PRESENTATION AND FEEDING METHOD ON DIETARY AME OF LAYING HENS

R.D. TAYLOR

Summary

A local and an imported strain of laying hens were subjected to complete or choice feeding regimes with calcium provided as limestone, either ground and included in the feed, or as grit (4-4.76 mm diameter) fed separately and provided daily or every second day. Dietary AME determined by a total collection method at 12, 20 and 29 weeks of age was higher ($P < 0.05$) for the local strain than the imported strain at 12 weeks but similar ($P > 0.05$) results were found at 20 and 29 weeks. Choice fed birds had a greater ($P < 0.05$) AME at 12 and 29 weeks than those given a complete diet but this result was reversed at 20 weeks. Similar ($P > 0.05$) dietary AME was found across calcium treatments at 12 weeks, but giving limestone grit either daily or every second day produced higher ($P < 0.05$) AME than ground limestone inclusion in the ration at 20 weeks. At 29 weeks AME was only improved ($P < 0.05$) by daily provision of limestone grit. Second daily provision of limestone grit resulted in a marked reduction of AME on the day of grit withdrawal. Providing coarse limestone grit to the imported bird improved dietary AME and this may be important where a small appetite causes nutrient intake to be marginal for maximum egg production.

I. INTRODUCTION

The issue of dietary calcium sources and levels for laying hens has been of considerable interest again in recent years. The NRC (1994) acknowledged that detailed research was required into the use of larger particle sizes of calcium. Apart from these larger particles providing calcium to the layer, there is the consideration of the use of these particles to aid in the grinding of feed in the gizzard of the bird. This subject has long been researched with great variation in results (McIntosh *et al.* (1962), Sibbald and Gowe (1977) and Cabrera *et al.* (1982)). Mollah (1982) attributed the possible improvement in AME caused by grits to the grinding action between grit and grain breaking the waxy surface coat of grains and thereby increasing the surface area on which the digestive fluids can act. However, in his own work Mollah (1982) found no effect on the AME of wheat when an insoluble grit of 2-4 mm dia. was fed to broilers. In laying hens, the provision of a calcium providing grit allows for the calcium requirements of the bird but may also act as an aid to improving dietary AME. Recent advice on the use of coarse calcium sources has recommended that it be as a portion of the total dietary calcium, which is an added burden for the diet compounder. Recommendations for substituting coarse for ground calcium sources needs to take account of the calcium provided in the diet to meet the daily calcium requirement, which is 3.6 g per bird for brown-egg layers (NRC, 1994).

Taylor (1996) had shown a non-significant reduction in the AME of a diet in laying hens when calcium was provided as shellgrit every second or fourth day compared with daily. It is not known whether this was due to changes in either calcium levels in the gut or to grinding effects with variation in grit levels in the gizzard.

Division of Animal Science, The University of New England, Armidale, NSW, 2351.

The present trial was undertaken to determine the effects, if any, of providing a calcium source solely as ground or particulate limestone on the AME of a diet presented in two forms to two strains of laying hens.

II. METHODS

One hundred and forty four commercial layers, of which half were an imported strain (HB) and half a local strain (CB), were introduced to either complete or choice feeding methods, using the same wheat-based formulation, from eight weeks of age. Within each feeding method calcium was provided as either; ground limestone included in the ration (Ca1); limestone grit (4-4.76 mm dia.) available daily in a separate feed trough (Ca2), or the same sized grit available every second day (Ca3). At 12 weeks of age three replicate pairs of birds per treatment combination were randomly selected from the rearing cages and placed in laying cages. Excreta were collected daily, for eight days, into aluminium foil trays suspended below the cage floors by wire. Excreta were bulked in two groups to allow for calcium treatment Ca3. In the laying house, at 20 and 29 weeks, three birds per treatment were randomly selected for 14 day excreta collections. Excreta were treated as per the rearing collection to allow for the birds receiving calcium treatment Ca3. AME was calculated from feed intakes and excreta output on a dry organic matter (DOM) basis.

Data from 12, 20 and 29 weeks were treated by factorial analysis in a 2 x 2 x 3 design within the GLM procedure of SAS. Differences between days, allowing for calcium treatment Ca3, were tested by repeated measures analysis within the GLM procedure of SAS. Relationships between AME versus either calcium and phosphorus intake, retention and percentage retention, and limestone grit intake was tested over strain, feed type and calcium method by the reduction in sums of squares technique (Snedecor and Cochran, 1980) using the GLM procedure of Minitab.

III. RESULTS

At 12 weeks of age (Table 1) dietary AME was higher ($P < 0.001$) in the CB than the HB strain and in the birds on choice feeding, rather than on the complete diet ($P < 0.01$). Providing calcium as either ground limestone or as particulate limestone daily or every second day had no effect ($P > 0.05$) on dietary AME. At 20 weeks (Table 1) there was no strain difference in AME ($P > 0.05$). Higher AME values were obtained with birds fed complete diets than when choice fed ($P < 0.01$). AME was lower ($P < 0.05$) when the calcium source was provided as ground limestone than as particulate limestone either daily or every second day. At 29 weeks (Table 1) there was no strain difference in AME ($P > 0.05$). AME was improved ($P < 0.001$) with the choice diet rather than the compound form. Providing calcium as ground limestone or as particulate limestone every second day resulted in a lower ($P < 0.001$) AME than providing particulate limestone daily.

A time effect was found at 29 weeks, when comparing AME allowing for days when calcium grit was presented or not to Ca3. A significant ($P < 0.05$) strain x form of dietary calcium interaction was found (Table 2). When limestone grit was provided every second day the AME of the diet was reduced on the day after access to the grit. This was more pronounced in the CB strain. The HB displayed a marked increase in dietary AME when the limestone was provided as grit.

Table 1. Least squares means for dietary apparent metabolizable energy (AME MJ/kg DOM) for HB and CB strain layers fed with a compounded or choice diet with calcium provided as ground limestone (Ca 1), limestone grit available *ad libitum* (Ca 2) or limestone grit available every second day (Ca 3).

Age (weeks)	Strain (n=18)		Feeding method (n=18)		Calcium method (n=12)		
	HB	CB	Complete	Choice	Ca1	Ca2	Ca3
12	14.61 ^b	15.00 ^a	14.68 ^b	14.94 ^a	14.76	14.73	14.93
SE	0.053		0.053		0.065		
20	15.10	15.12	15.24 ^a	14.97 ^b	14.93 ^b	15.27 ^a	15.13 ^a
SE	0.055		0.055		0.068		
29	14.73	14.77	14.62 ^b	14.88 ^a	14.53 ^b	15.04 ^a	14.69 ^b
SE	0.070		0.070		0.085		

^{ab} Means with unlike superscripts within factors and ages are significantly different (P<0.05).

Table 2. Least squares mean for dietary AME (MJ/kg DOM), at 29 weeks, for HB and CB strain layers given dietary calcium by the three methods.

AME MJ/kg DOM	HB			CB			SE
	Ca1	Ca2	Ca3	Ca1	Ca2	Ca3	
Grit	14.48 ^a	15.24 ^c	14.85 ^b	14.67 ^{ab}	14.82 ^{ab}	14.98 ^{bc}	0.128
No Grit	14.24 ^a	15.15 ^d	14.45 ^{ab}	14.72 ^{bc}	14.93 ^{cd}	14.50 ^{ab}	0.136

^{abcd} Means with unlike superscripts within a row are significantly different (P < 0.05).

Significant relationships at 29 weeks between dietary AME and calcium intake and phosphorus retention and percentage retention across strain, feed or calcium method are shown in Table 3.

Table 3. Relationships at 29 weeks between AME (Y; MJ/kg) and Ca intake or P retention (X; g/kg body weight) of HB and CB strain birds fed a complete or choice diet with Ca provided by the three methods.

AME vs	Factor	Regression Equation	R ²	P ¹	RSD
Ca intake (g)	Complete	Y = 14.597 + 0.009 X	0.001	NS	0.31
	Choice	Y = 14.139 + 0.404 X	0.191	*	0.40
P retention (g)	HB	Y = 14.070 + 8.151 X	0.702	***	0.27
	CB	Y = 14.697 + 0.800 X	0.001	NS	0.29
P retention (%)	HB	Y = 13.853 + 0.044 X	0.734	***	0.25
	CB	Y = 14.551 + 0.010 X	0.024	NS	0.28
P retention (%)	Ca1	Y = 14.461 + 0.004 X	0.001	NS	0.32
	Ca2	Y = 13.756 + 0.049 X	0.786	***	0.20
	Ca3	Y = 14.115 + 0.029 X	0.484	**	0.20

¹ * = P<0.05; ** = P<0.01; *** = P<0.001; NS = P>0.05.

IV. DISCUSSION

AME at 12 weeks and peak lay was 99.8 and 99.4% of that prior to point of lay (20 weeks) in choice fed birds but 96.3 and 95.9% in birds given compounded feed. AME increased as energy intake fell (data not presented) at point of lay especially in complete fed birds. This was altered at peak lay when the second-daily provision of limestone grit resulted in a similar AME to that from ground limestone whilst daily grit caused a greater energy utilisation. The complex interaction of strain and method of calcium provision with AME at peak lay provides one reasonably clear result, whereby providing limestone grit every second day caused a large change in AME between days when grit was available or not. In the HB strain this was -0.401 MJ and in the CB strain was -0.482 MJ. As the grit was the major calcium source, with grit supplied every second day, normal digestive activity may have been adversely affected either by a lack of calcium *per se* or by the rapid transit of the particulate calcium source. The significant relationship between AME and calcium intake for the choice fed birds supports the above. However, calculation revealed that these birds were eating 3.66 g/d of calcium, based on mean body weights of 2 kg, which matches the NRC (1994) recommendations of 3.6 g/d of calcium for brown-egg layers and exceeds the 3.4 g/d set by Leeson *et al.* (1993) as the maximum requirement of brown-egg layers. These recommended levels may be inadequate for some heavy laying strains.

Strong relationships between AME and P parameters are indicated at peak egg production. In the HB bird dietary AME was improved ($R^2 = 0.702$) with greater P retention (g/kg body weight) and this was reinforced by the P % retention ($R^2 = 0.734$). To attain peak dietary efficiency, the HB strain may require a higher dietary available P level. The birds offered daily calcium grit retained more P than from the other two calcium methods, irrespective of similar intakes (data not presented). It is suggested that giving limestone grit daily to laying birds may allow the phytate P in the wheat to be rendered more available to the bird which in turn allows an improved dietary AME. The effect of limestone grit intake on AME results was not clearly elucidated and detailed work is underway to determine if the effect was due to grinding of the feed, whether already milled or not, due to an improvement in calcium availability in the lower gut, or a combination of both.

V. ACKNOWLEDGMENTS

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ACUTE VITAMIN D DEFICIENCY IN LAYERS

R.B. CUMMING¹, B. FIELD² and V. FIELD²Summary

The initial problem was observed in a flock of 5,000 40-week-old layers, many unable to stand, in the middle of an Egg Drop Syndrome (EDS) outbreak. A deficiency of calcium was diagnosed. The flock was then moulted. About four weeks later, a second flock of 10,000 young layers, which had peaked at 92% production, crashed to about 20% in two weeks. These birds were housed in a saw-toothed shed, with 18 rows each of about 550 birds. Every fourth row received some direct sunlight and the production in these rows had fallen to about 40%. Rows that received no sunlight fell to as low as 3%.

A diagnosis of vitamin D₃ deficiency was made. Some rows of birds received water dispersed A, D and E over their crumbles, while other rows were placed on feed from a different feed mill. Within 72 hours, production improved, as did egg shell quality.

The first flock had been brought back into production but reached only about 50% before collapsing. Gross lesions of vitamin D₃ deficiency were observed. Following dietary Vitamin D₃ supplementation, production in these flocks increased to 88%. The implications of this problem, which is obviously widespread and not being diagnosed, will be discussed.

I. INTRODUCTION

Vitamin D₃ is normally incorporated in modern poultry diets as a stabilised powder and there are no reports of acute vitamin D₃ deficiency in modern layers in the literature. Very abrupt drops in egg production in modern flocks are often associated with infectious disease agents such as Avian Encephalomyelitis virus, Infectious Bronchitis and, more recently, Egg Drop Syndrome (EDS) virus. This report details the findings on farms where EDS had occurred, but the picture was confounded by a concurrent acute Vitamin D₃ deficiency.

II. DESCRIPTION OF PROBLEM

The farmer concerned has several properties housing some thousands of laying hens in conventional three-bird cages in saw-toothed sheds. In addition, he rears large numbers of point-of-lay pullets, some on the floor and some in wire floored cages. All the birds are fed on crumbled rations (chicken starter, pullet grower and layer) produced by a large feed firm.

Egg-Drop Syndrome infections were recorded in the Tamworth area in March 1997. The initial problem was observed in April 1997 in a 5000-bird shed of 40-week-old layers (flock A) with birds dying for no apparent reason in the middle of an EDS outbreak.

On investigation, about 5% of the layers had died and a number of birds were unable to stand in the laying cages. These were removed and placed on the ground. Most were incapable of walking but otherwise appeared normal. Autopsies were then carried out on 24 birds that had died the previous day. Two birds were affected with Marek's Disease, while the remainder were all in full production and apparently healthy. Generally these birds appeared to have been suffocated with marked engorgement of the blood vessels on the ovaries and oviducts.

¹ Department of Animal Science, University of New England, Armidale, NSW 2351.

² "Wyoming", Moonbi NSW 2353.

The ribs of the birds were observed to be very soft and easily cut with dissecting scissors. The tibias broke fairly readily, with most of them showing a classical green-stick fracture.

On returning to the laying shed where the birds had been placed on the ground, approximately half of these birds had laid eggs. These hens were now eating broken egg shells lying underneath the laying cages. A tentative diagnosis of calcium deficiency was made, the feed firm advised, and oyster shell chips dispersed over the food in the feeders. Instructions were left to remove birds with leg problems in the cages at least twice daily and to place them on the floor with suitable feed and water nearby.

The owner advised a week later that the mortality had ceased, and that the birds placed on the ground had stopped laying but regained the use of their legs and appeared normal. These were then returned to the laying cages and the flock was being moulted because of the impact of the EDS which had been confirmed serologically. In addition, the feed was being tested for calcium content. The owner then vaccinated all of his other laying flocks against EDS. Two weeks later the owner was advised that the ration was adequate in calcium at 37g/kg.

About four weeks later at a second farm, 10,000 young brown-egg layers (flock B) which had peaked at 92% production by 23 weeks of age, crashed to about 60% production in ten days. This farm was visited a few days later when egg production had dropped to 30%. Egg shell quality was extremely poor, and a few birds were unable to stand. These birds were housed in a saw-toothed shed, three birds to a cage, with eighteen rows of approximately 550 birds each. It was noticed that every fourth row in the shed received direct sunlight through the openings of the saw-toothed shed which faced north. The rows (numbers 4, 8, 12 and 16) receiving direct sunlight had fallen to about 40% production, while rows that received no sunlight fell as low as 9% (see Table 1). A number of dead birds were autopsied and these showed the same soft bones as seen on the first property (flock A).

A diagnosis of vitamin D₃ deficiency was made, and the feed firm advised, but the diagnosis was contested.

III. TREATMENTS AND RESPONSES

It was therefore decided to place some rows on feed from a different feed mill and when the materials were received, to supplement some rows of birds with water-dispersed vitamins, A, D₃ and E over their feed. The remaining rows were maintained as untreated controls.

As shown in the table, within 72 hours the rows receiving the diet from the second feed mill (rows 8, 9 and 10), or water-dispersed vitamin D₃ (rows 1, 2, 3 and 4) improved in production. Egg shell quality improved rapidly and, within ten days, these rows increased to about 60% production. The rows that had not received additional vitamin D or the new diet remained at their previous levels of production.

As more water dispersed fat soluble vitamin mixtures became available, additional rows were supplemented or placed on a new diet with adequate vitamin D₃. All showed the same steady rise in production after 72 hours.

The first flock which had experienced the EDS outbreak had been rested for several weeks and then brought back into production. These birds climbed swiftly to about 50% production before collapsing again with egg production dropping dramatically and large numbers of birds showing leg problems and dying. Gross lesions of vitamin D₃/calcium deficiency were observed in these layers.

Two additional flocks of approximately 10000 birds, each of which had been in production for about 50 weeks of age, had been producing extremely poorly, with a large number of broken eggs. The owner thought they lacked persistency and was thinking of

moulting these birds. These birds also showed green stick fractures. On the rearing farm ten pullets from each of two groups aged six and ten weeks were killed and examined for symptoms of vitamin D₃ deficiency. None was observed, the bones breaking quite cleanly.

The feed firm now agreed that the problem lay with the feed and all the birds on the second farm were placed on new rations with a new source of vitamin D. The production on this second farm (flock B) reached a consistent 88% ten days later.

IV. DISCUSSION

The response of the young hens (flock B) to the addition of fat soluble vitamins to their diet was quite dramatic, as was the response to placing the birds on a diet adequate in vitamin D₃. Within 72 hours, production started to increase, reaching about 50% in five days and up to 80% in ten days. Of the rows maintained on the original diet and not supplemented until 3 July, at that time the row with sunlight (row 12) was still at approximately 32% production while the other unsupplemented rows (rows 5, 6, 11, 13, 14 and 15) were at approximately 7% production.

The fact that the growing birds, also fed on pelleted or crumbled rations, and not showing gross lesions of vitamin D₃ deficiency, suggests that the vitamin D₃ in the rations was marginal. There was sufficient of the vitamin for egg-type growing stock but insufficient for laying birds. Nutritional recommendations, e.g. Scott *et al.* (1976), suggest that laying birds require at least double and sometimes quadruple amounts of vitamin D₃, compared to growing birds. The feed firm concerned, which had commenced using a new large consignment of vitamin D₃ concentrate in the middle of March, supplies vitamin premixes to a large number of poultry farmers and feed mills in eastern Australia, as well as supplying many pig producers with feeds in pellet or nut form. While no other complaints of a vitamin D₃ deficiency in layers had been made, most laying hens are fed on mash rather than pelleted rations and thus the premix and the fat-soluble vitamins in particular, would not have been exposed to the deleterious effects of high temperature in the pelleting process.

In general, the vitamin D₃ recommendations for pigs are essentially similar to that for growing egg-type stock. A large local turkey farm was supplied with crumbled and pelleted diets by the same feed firm but reported no unusual leg problems. However, investigation showed that all the turkeys on this property had access to direct sunlight from day-old.

Very few other egg producers in the Tamworth area used feed from this mill. However, one farm of approximately 15,000 layers situated near Port Macquarie, did use the same crumbled layer diet and this farmer had recently purchased point-of-lay pullets from the Tamworth farmer concerned. The pullets crashed in egg production in a similar fashion to Tamworth flock B. A diagnosis of EDS was made by this farmer but what he found puzzling was that half the pullets, which were fed on a crumbled layer diet from a different feed mill, continued to produce extremely well. Treatment with vitamin D rapidly restored his affected flock to normal production.

At the time of this problem, considerable difficulty was experienced in obtaining the water-dispersed mixtures of vitamins A, D₃ and E. Apparently, many producers had found there was a benefit to be gained by administering these vitamins to their layers and broilers. Such treatments, while highly unscientific, may partially account for the fact that other flocks did not go down with the frank symptoms of vitamin D₃ deficiency.

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Table. The effects of vitamin D deficiency and subsequent treatments on the rate of lay (%) over 21 days of 9,600 young layers housed in 18 rows of cages in a laying shed.

Row	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	Total	Treatment	
Bird No.	559	563	552	552	561	563	545	551	562	564	542	546	540	535	552	569	548	194	9598		
Day	Date																				
1	19/6	10.9	34.9	13.9	32.9	27.3	16.2	11.0	50.8	21.5	9.2	8.5	26.6	22.4	12.7	13.0	52.7	64.8	30.3	26.0	
2	20/6	6.8	31.4	4.3	29.9	20.3	13.1	6.4	43.9	15.6	7.1	6.5	38.1	17.4	10.8	10.3	42.7	54.0	57.7	21.8	Rows 8, 9 & 10 changed to alternative feed.
3	21/6	6.3	22.4	8.9	27.2	13.7	11.4	6.2	45.0	17.1	5.8	7.4	33.1	15.2	8.6	7.8	43.1	41.8	50.5	19.4	
4	22/6	7.8	29.8	11.6	28.1	16.2	9.8	5.7	60.1	35.2	18.1	6.1	44.5	21.5	10.3	12.8	45.6	48.7	52.5	24.9	
5	23/6	10.9	23.1	10.9	25.4	15.2	6.7	5.9	58.3	39.9	32.9	6.4	40.1	15.2	8.0	6.2	39.2	34.7	43.3	22.6	Vit A,D,E over feed in rows 1-4. Row 7 to alternative feed.
6	24/6	5.5	19.7	11.8	26.8	10.8	6.6	6.2	66.2	50.4	36.5	5.4	38.2	15.0	6.4	6.3	40.6	29.7	41.2	22.9	
7	25/6	15.4	28.4	19.9	37.8	11.0	6.4	22.2	70.0	55.6	46.8	5.9	36.8	14.3	6.4	7.8	37.3	29.3	34.5	26.8	
8	26/6	23.6	34.8	29.3	42.5	12.2	3.9	27.1	68.6	59.3	48.2	4.8	36.1	13.5	7.7	4.9	36.0	22.8	33.5	28.2	Vit A,D,E over feed in rows 1-4.
9	27/6	29.1	39.2	39.1	54.3	11.6	6.0	39.8	77.3	68.7	63.5	5.2	34.9	16.1	11.0	8.3	39.5	19.9	29.9	33.2	
10	28/6	43.8	53.3	53.3	58.5	11.8	5.3	45.1	76.0	63.2	63.4	4.8	37.4	18.9	9.3	4.9	42.2	20.6	42.8	36.1	Row 18 changed to alternative feed.
11	29/6	49.4	58.1	59.2	66.3	17.5	6.2	56.5	77.7	72.1	65.1	4.8	34.8	20.2	6.2	6.3	41.8	17.1	42.7	39.0	
12	30/6	50.9	53.5	57.9	64.3	14.8	6.0	54.3	80.2	79.5	69.7	3.3	33.2	20.7	8.9	5.4	42.4	16.6	51.5	39.4	
13	1/7	62.1	68.4	71.0	73.2	19.3	4.3	67.3	84.4	78.6	63.3	4.8	37.4	21.1	5.6	5.3	44.1	18.4	58.2	43.3	
14	2/7	59.4	65.5	71.6	68.8	17.6	4.3	58.9	76.8	72.9	68.6	5.4	28.6	16.9	5.4	5.4	36.2	17.2	57.7	40.4	
15	3/7	58.5	63.4	68.3	66.3	16.0	4.4	68.1	82.9	74.9	68.8	4.2	31.7	15.4	4.9	4.9	34.6	15.1	59.8	40.7	Reformulated original feed to rows 1-6 and 11-17.
16	4/7	66.5	71.8	76.6	85.9	22.8	5.3	66.9	81.1	81.1	76.1	6.5	38.5	15.7	6.5	4.3	42.5	18.9	76.8	45.9	
17	5/7	71.0	72.1	78.4	88.4	32.9	15.5	73.4	84.8	82.7	73.8	17.3	47.4	33.5	11.2	18.5	49.2	22.4	80.9	52.1	Vit ADE supplement to rows 1-16, excluding No 6 (just D)
18	6/7	66.4	65.5	73.2	71.4	40.1	26.8	67.5	82.0	71.9	80.1	30.9	52.4	44.6	26.9	27.5	53.9	31.0	71.6	54.1	This was repeated on day 18.
19	7/7	61.4	73.0	75.7	77.4	47.2	36.9	71.4	78.4	75.9	69.3	45.8	63.6	60.7	47.7	46.9	72.9	48.5	81.9	62.3	
20	8/7	70.7	67.9	77.5	77.9	65.8	55.8	69.7	90.7	82.2	79.4	35.2	73.4	47.6	53.6	71.0	74.9	53.5	75.8	67.7	
21	9/7	79.6	79.4	84.2	81.3	63.1	61.6	74.7	83.1	83.8	85.6	55.9	67.8	62.3	63.9	62.1	68.5	65.5	79.8	72.4	

THE EFFECT OF DIFFERENT XYLANASES ON CARBOHYDRATE DIGESTION AND VISCOSITY ALONG THE INTESTINAL TRACT IN BROILERS

M. CHOCT

Summary

The mode of action of two xylanase products on the digestion of non-starch polysaccharides (NSP) and digesta viscosity along the gut was examined in broilers fed two wheats with different metabolisable energy values. Significantly ($P < 0.01$) higher viscosity values (mPa.s) were observed in the duodenum (3.68 vs 2.58), jejunum (5.14 vs 2.6) and ileum (18 vs 10.6) of birds fed the low-ME wheat compared with those fed the normal wheat. In the duodenum, both enzyme products lowered ($P < 0.05$) the viscosity regardless of wheat type, but in the jejunum, a reduction in viscosity was observed only in the birds fed the low-ME wheat. Both enzymes caused an increase in the ileal viscosity. The relative level of soluble NSP in the ileum was significantly ($P < 0.01$) elevated by both enzymes and this coincided with a decrease in the insoluble NSP level. The enzymes did not release substantial amounts of monosaccharides and oligosaccharides.

I. INTRODUCTION

The extreme variation in the apparent metabolisable energy (AME) value of wheat in poultry is related mainly to the level of soluble non-starch polysaccharides (NSP), which elicit anti-nutritive activities, depressing nutrient digestion and absorption (Annison, 1991). Extensive studies have been conducted to elucidate the mechanism of the anti-nutritive activities of soluble NSP in order to find effective solutions for the problem. One of the solutions is the use of microbial enzyme supplements. It is, however, not without its share of problems. The commercial enzyme products vary in their efficacy for performance enhancement as well as other attributes in monogastric diets. This stems from the difficulties in accurate quantification of the enzyme activities and poor understanding of the substrates.

In the current study, the effect of two enzyme products on various carbohydrate fractions and digesta viscosity along the gut was studied in broilers fed two wheats with different AME values.

II. MATERIALS AND METHODS

Trial diets: Two wheats with known AME values were chosen from a survey for this study. The diets consisted of the two wheats with and without two xylanase products and a maize control. The enzyme products were added at an equivalent of 5600 xylanase units per kg of diet. The Sydney University AME basal diet was used in this study (Mollah *et al.*, 1983). Celite (Celite Corporation, Lompoc, California, USA) was added as a digesta marker. All ingredients were thoroughly mixed in a small mixer and cold-pelleted. Each diet was fed to 5 replicates of 4 birds each.

AME trial: Inghams AM98 broilers (300 birds), 27 d of age, were weighed and 140 healthy birds of similar liveweight were chosen. These birds were transferred to metabolism cages in an insulated room maintained at 22-26°C and were fed the trial diets for 7d.

School of Rural Science and Natural Resources - Division of Animal Science, University of New England, Armidale, NSW 2351.

The first three days served as an adaptation period and for the last four days excreta were collected daily, dried overnight in a forced-air oven at 80°C and pooled for determination of gross energy. Feed intakes were measured for the entire 7-d period and at the end of the trial bird weights were recorded for calculation of growth rate and feed conversion ratio.

Collection of digesta: At the end of the AME trial two birds were randomly chosen from each cage and killed by intravenous injection of Nembutal. The contents of the crop, duodenum, jejunum and ileum (10 cm from Meckel's diverticulum to 1 cm above the ileo-caecal junction) were collected and spun at 12,000 g for 15 min. The supernatant was taken and stored at -20°C until viscosity was determined. Freezing had no effect on viscosity as demonstrated by an ancillary trial. Measurements on the crop contents were abandoned due to difficulty in obtaining sufficient crop contents from many of the birds.

Viscosity measurement: Viscosity was determined with 0.5 mL supernatant using a Brookfield viscometer at 25°C with a CP40 cone and shear rate of 5-500 s⁻¹. The samples did not exhibit shear thinning at these shear rates. After the determination of the ileal viscosity, the supernatant was re-combined with the solid and freeze-dried for nutrient analyses.

Acid-insoluble ash (AIA): Diet (2-3 g) and digesta (1 g) samples were weighed into sintered crucibles (porosity 4) and dried at 105°C for 6 h. They were then accurately weighed, ashed (480°C, 8 h), and treated twice with boiling 4 M HCl. The residue was washed, dried (105°C, 6 h) and collected as the acid insoluble ash. The small intestinal digestibility coefficient (DC) of nutrients was calculated using the following equation:

$$DC = 1 - \frac{\text{Ileal nutrient (g/kg)} / \text{Ileal AIA (g/kg)}}{\text{Diet nutrient (g/kg)} / \text{Diet AIA (g/kg)}}$$

Gross energy: Diet and excreta gross energy was determined using an isoperibol Parr bomb calorimeter (Parr Corporation, USA).

Free sugars and soluble and insoluble non-starch Polysaccharides: The levels of free sugars, and soluble and insoluble NSP of the ileal contents were determined using the Uppsala method (Theander and Westerlund, 1993). Ileal contents (100 mg) were extracted twice with diethyl ether (5 mL) (to remove fat and pigments) and centrifuged at 2000 g for 15 min. The supernatants were discarded. The residues were then extracted with 80% ethanol and centrifuged (2000 g, 15 min). The supernatants were taken, dried under N₂ and hydrolysed in 1 M H₂SO₄ at 100°C for 2 h for determination of the free sugar content. The sample residues were treated with thermostable α-amylase and amyloglucosidase to remove starch residues. Then they were hydrolysed, reduced and acetylated as described by Englyst and Hudson (1993). The NSP in digesta were expressed as g NSP per kg AIA.

Statistical analyses: Firstly, the data were analysed as a 2 x 2 factorial design (two wheats and two enzymes) excluding the maize control. Then the data were analysed using one-way analysis of variance and multiple comparisons were based on the least significant difference test. Statgraphics (STSC Inc, Rockville, MD, USA) was used to perform the analyses.

Animal Ethics: The animal experiment was conducted in South Australian Research and Development Institute in Adelaide. The Animal Ethics Committee of the Department of Primary Industries (South Australia) approved this study. Health and husbandry practices

compiled with the "Code of Practice for the Welfare of the Domestic Fowl" issued by the Australian Bureau of Animal Health in 1983.

III. RESULTS

There were no significant interactions between wheat type and enzymes for any of the parameters measured. Both enzymes significantly improved ($P<0.01$) weight gain, feed conversion efficiency of birds fed the low-ME wheat. The AME of the low-ME wheat was also significantly ($P<0.05$) improved by both enzymes. Supplementation of the low-ME wheat diet with xylanase B increased ($P<0.05$) feed intake of the birds. No other significant effect of was observed on feed intake. Both enzymes had no significant effect on nutritive value of the good quality wheat (Table 1). Markedly higher viscosity values (mPa.s) were observed in the duodenum, jejunum and ileum in the birds fed the low-ME wheat diet compared with those fed the normal wheat diet. In the duodenum, both enzymes lowered the digesta viscosity of birds regardless of wheat type, but it was significant ($P<0.05$) only in birds fed the low-ME wheat. In the jejunum a trend of reduction in viscosity was observed in the birds fed the low-ME wheat. The two enzyme products tended to increase the ileal viscosity of birds fed both wheats, with Enzyme B markedly ($P<0.05$) elevating the digesta viscosity of birds fed the low-ME wheat (Table 2). Soluble NSP in the ileum were significantly ($P<0.01$) elevated by the supplementation of the enzyme products and this was related to decreases in the insoluble NSP. The enzymes did not release substantial amounts of monosaccharides and oligosaccharides (Table 3).

Table 1. Effects of two different xylanases on weight gain (WG; g/bird/week), feed intake (FI; g/bird/week) and feed conversion ratio (FCR; feed: gain) of broilers fed normal and low-ME wheat diets. Apparent metabolisable energy (AME; MJ/kg dry matter) values are also shown (n=5; means \pm SE).

Diet	WG	FI	FCR	AME
Maize control	448 \pm 14 ^a	865 \pm 19 ^{ab}	1.935 \pm 0.035 ^a	15.59 \pm 0.15 ^a
Low-ME wheat	379 \pm 41 ^b	792 \pm 46 ^b	2.144 \pm 0.138 ^b	12.70 \pm 0.39 ^b
Low-ME wheat + xylanase A	449 \pm 11 ^a	854 \pm 19 ^{ab}	1.908 \pm 0.061 ^a	14.02 \pm 0.41 ^c
Low-ME wheat + xylanase B	488 \pm 15 ^a	893 \pm 19 ^a	1.834 \pm 0.043 ^a	13.81 \pm 0.38 ^c
Normal wheat	435 \pm 20 ^{ab}	827 \pm 29 ^{ab}	1.904 \pm 0.027 ^a	14.30 \pm 0.13 ^c
Normal wheat + xylanase A	434 \pm 18 ^{ab}	817 \pm 27 ^b	1.886 \pm 0.036 ^a	14.32 \pm 0.35 ^c
Normal wheat + xylanase B	478 \pm 9 ^a	858 \pm 12 ^{ab}	1.795 \pm 0.014 ^a	14.26 \pm 0.14 ^c

abc Means with unlike superscripts within a column differ significantly at $P<0.05$.

Table 2. Effects of two different xylanases on gut viscosity (mPa.s) of broilers fed normal and low-ME wheat diets (n=5; means \pm SE)¹.

Diet	Duodenal Viscosity	Jejunal Viscosity	Ileal Viscosity
Maize control	1.54 \pm 0.06 ^a	2.22 \pm 0.10 ^a	3.80 \pm 0.39 ^a
Low-ME wheat	3.68 \pm 0.61 ^b	5.14 \pm 0.83 ^b	18.14 \pm 3.42 ^{bcd}
Low-ME wheat+Xylanase A	1.92 \pm 0.29 ^{ac}	3.56 \pm 0.84 ^{abc}	22.40 \pm 3.42 ^{bcd}
Low-ME wheat+Xylanase B	2.08 \pm 0.10 ^{ac}	3.94 \pm 0.69 ^{bc}	27.42 \pm 4.87 ^d
Normal wheat	2.58 \pm 0.24 ^c	2.60 \pm 0.13 ^{ac}	10.62 \pm 2.21 ^{ab}
Normal wheat+Xylanase A	2.46 \pm 0.32 ^c	2.74 \pm 0.41 ^{ac}	16.58 \pm 3.34 ^{bc}
Normal wheat + Xylanase B	1.82 \pm 0.10 ^{ac}	2.20 \pm 0.30 ^a	14.70 \pm 3.69 ^{bc}

abcd Means with unlike superscripts within a column differ significantly at $P<0.05$.

Table 3. Effects of two different xylanases on soluble and insoluble non-starch polysaccharide (NSP) levels and free sugars (g/kg of acid-insoluble ash) in the ileum of broilers fed normal and low-ME wheat diets (n=5; means \pm SE).

Diet	Soluble NSP	Insoluble NSP	Free Sugars
Maize control	92 \pm 7 ^a	2629 \pm 118 ^a	188 \pm 26 ^a
Low-ME wheat	413 \pm 31 ^{bc}	2773 \pm 109 ^{bc}	401 \pm 125 ^{ad}
Low-ME wheat + Xylanase A	572 \pm 47 ^d	2400 \pm 84 ^a	493 \pm 77 ^b
Low-ME wheat + Xylanase B	749 \pm 51 ^e	2495 \pm 46 ^a	413 \pm 99 ^{ab}
Normal wheat	321 \pm 30 ^b	2864 \pm 152 ^c	348 \pm 58 ^{ab}
Normal wheat + Xylanase A	496 \pm 71 ^{cd}	2458 \pm 146 ^{ab}	611 \pm 150 ^b
Normal wheat + Xylanase B	479 \pm 33 ^{cd}	2324 \pm 107 ^a	414 \pm 47 ^{ab}

abcde Means with unlike superscripts within a column differ significantly at P<0.05.

IV. DISCUSSION

Supplementation of a diet containing a low-ME wheat with two different xylanases markedly improved its nutritive value for broiler chickens, increasing weight gain and feed conversion efficiency. No marked improvement was seen in birds fed the normal wheat. The normal wheat diet used in the current experiment had a nutritive value close to that of the maize control diet and therefore a substantial improvement of its nutritive value was not expected.

The quality and quantity of the NSP in wheat vary due to environmental and climatic conditions under which the wheat is grown (Choct *et al.*, 1996) and the soluble NSP level is inversely related to the AME in wheat (Annison, 1991). In the current experiment, the digesta viscosity along the GI tract was much higher in birds fed the low-ME wheat diet than those fed the normal wheat diet and it was inversely correlated to bird performance. In the chicken, when the digesta reaches the ileum, most nutrients are digested with NSP remaining to make up the bulk of ileal contents. This was clearly demonstrated in the current experiment, which showed a progressive increase in digesta viscosity along the GI tract with values many fold higher in the ileum than in the duodenum. Both enzymes effectively reduced digesta viscosity in the duodenum and jejunum. On the contrary, both enzymes markedly increased the ileal viscosity. This, however, had little effect on the overall performance of the birds. In chickens, most nutrients are digested in the upper part of the digestive tract and reduction of digesta viscosity in the duodenum and jejunum therefore appears to be important for the alleviation of the anti-nutritive activity of soluble NSP in poultry diets. The elevation of the ileal viscosity appeared to be the result of solubilisation of the insoluble NSP by the enzymes. Glycanases vary widely in their substrate affinity. Whilst some have strong affinity for the soluble NSP to reduce gut viscosity, others have affinity for both soluble and insoluble NSP (Gruppen *et al.*, 1993). The xylanases used in the current experiment appear to belong to the latter category. Solubilisation and partial breakdown of insoluble NSP may encourage increased fermentation of the NSP in the hindgut, which in turn could enhance energy availability to the bird via production of volatile fatty acids.

No substantial amounts of monosaccharides and oligosaccharides were detected in the ileal contents. Whether this was due to an efficient disappearance of the released sugars in the gut or due to the inability of the enzymes to release free sugars is not known, but it is generally conceded that the improvement of the nutritive value of cereal-based diets by enzymes is not due to a complete cleavage of the NSP and a subsequent absorption of the released sugars. It is due to the ability of the enzymes to partially depolymerise the soluble NSP to eliminate their anti-nutritive effects on nutrient digestion and absorption.

It may be concluded that reduction of digesta viscosity in the duodenum and jejunum of chickens fed viscous grains is an important attribute of feed carbohydrates.

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OPTIMISING THE DOSE OF XYLANASE IN BROILER DIETS BASED ON WHEAT QUALITY AND ECONOMIC FACTORS

M. PACK¹, M. BEDFORD¹ and D. CRESWELL²

Summary

The present paper outlines a model developed to determine the most economical xylanase dose rates for use in broiler diets based on wheat. In this model, wheat quality and the subsequent response potential to xylanase addition are estimated via *in vitro* extract viscosity determination in the wheat grain. The extract viscosity data correlated closely with *in vivo* digesta viscosity in the bird. Feed conversion ratio (FCR) response prediction to graded enzyme doses provides a basis for calculation of marginal income versus marginal cost, which can then be used to determine the economic dose optimum for any given situation.

I. INTRODUCTION

The application of xylanase-based enzymes in wheat diets for broilers has become a routine practice in countries where wheat is the most cost-effective cereal. It is recognised that the high viscosity of the intestinal digesta brought about by the soluble arabinoxylans present in wheat is the major problem in these diets. Endo-xylanases are an effective means of reducing viscosity by partially breaking down these non-starch polysaccharide components (Choct and Annison, 1992; Bedford and Morgan, 1996; Smits and Annison, 1996). It has also been shown that differences in the feeding value of different wheats for broilers closely correlate with the *in vivo* digesta viscosity in the chicken gut. Increasing viscosity due to increased content of soluble arabinoxylans will lead to lower wheat AME content and higher FCR in broilers (Classen *et al.*, 1995; Jeroch *et al.*, 1997; Barrier-Guillot *et al.*, 1997).

For the practical application of this knowledge, however, there is a need to develop an *in vitro* tool to conveniently determine wheat quality on small samples, and then use this information to predict responses to xylanase addition. The present paper outlines a model integrating the *in vitro* estimation of wheat quality with response prediction to graded levels of xylanase supplements, where the enzyme dose optimum is determined based on the economics of the responses relative to the cost of the enzyme.

II. EXTRACT VISCOSITY CHARACTERIZES WHEAT QUALITY

Various attempts have been made to establish an *in vitro* method which accurately predicts *in vivo* intestinal viscosity. However, they have generally failed to give an accurate reflection of the *in vivo* result. These methods were adequate to distinguish between very good and very poor quality wheat, but did not always give a close fit to the full range of viscosities seen in the chick assay and consequently in bird performance (Classen *et al.*, 1995; Jeroch *et al.*, 1997). In our laboratory, we have recently developed a more sophisticated approach to this assay by introducing *in vitro* enzymatic digestion steps before extracting the soluble fibre components and measuring viscosity of the extract (aviCheckTM extract viscosity, Finnfeeds proprietary method, unpublished).

¹ Finnfeeds International Ltd, Marlborough SN8 IXN, United Kingdom.

² Creswell Livestock Consultants, Mosman, NSW 2088.

This method has been shown to give a good fit to the digesta viscosity¹ measured *in vivo*, and hence provides the tool to determine wheat (and other viscous cereals) quality in a rapid and comparatively inexpensive laboratory assay (Figure 1).

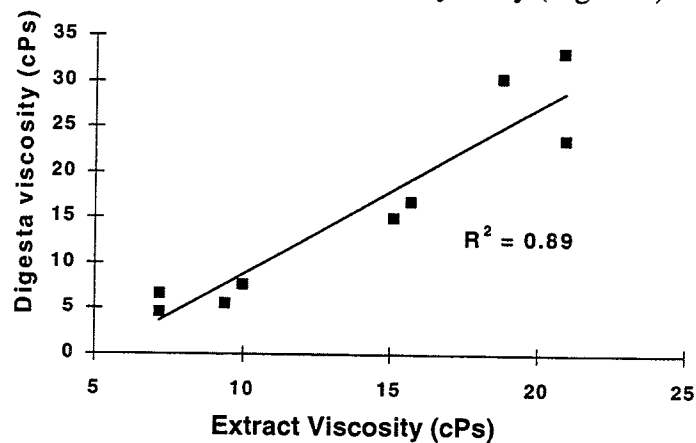


Figure 1. Correlation between digesta viscosity and *in vitro* wheat extract viscosity.

III. PREDICTING THE XYLANASE RESPONSE POTENTIAL

The next step towards establishing the most economical xylanase dose rate in wheat diets requires a good prediction of the maximum response potential (expressed as broiler FCR or wheat AME) to xylanase addition. It has been shown that the response will depend on the initial quality of the cereal in the diet. In fact there is a strong linear correlation between the wheat extract viscosity and the AME in young broilers, and the response to xylanase is larger in the low AME wheats (Figure 2). It is apparent, therefore, that the determination of extract viscosity provides the most important measure of wheat quality in order to estimate how much response to enzyme addition can be expected. There are a few other factors that have been shown to have an impact on the size of the enzyme response, such as dietary fat type (Dänicke *et al.*, 1995; Choct *et al.*, 1996), wheat inclusion level or processing temperature (Nissinen, 1994). These factors are worth accounting for in response prediction, but it appears that viscosity is by far the most important single predictor of the enzyme response in wheat diets for broilers (Bedford and Morgan, 1996; Barrier-Guillot *et al.*, 1997).

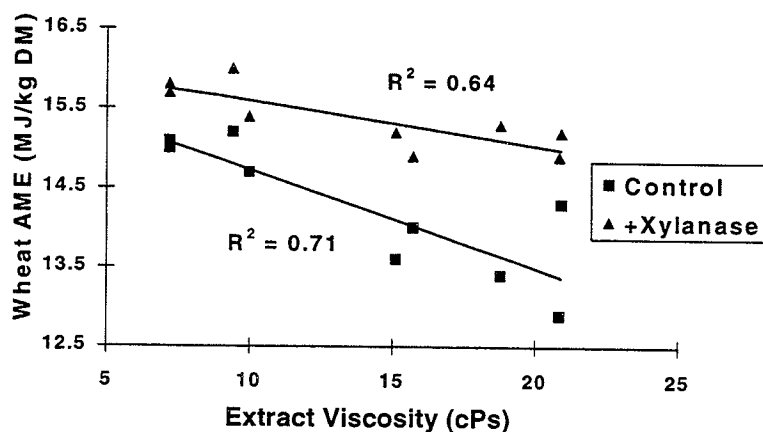


Figure 2. Correlation between wheat AME and *in vitro* extract viscosity (AME data as determined by Classen *et al.* (1995) vs. aviCheckTM wheat extract viscosity).

¹ aviCheckTM, trademark Finnfeeds International Ltd

The enzyme used in the trials referred to in this paper was Avizyme 1300 (*Trichoderma longibrachiatum* xylanase, 2500 U/g, *Bacillus subtilisin* protease, 800 U/kg). Dose rate was held constant at 1 kg per tonne. Although xylanase addition alleviated much of the initial variability in wheat AME, there is still a difference between the best and the worst wheats even after enzyme addition. This has been demonstrated for both wheat and barley (Hughes *et al.*, 1996; Kocher *et al.*, 1997), and raises the questions (1) whether higher enzyme dosage would have been beneficial for the highly viscous wheats, and (2) whether the full dose of 1 kg/tonne is necessary for the wheats showing lower viscosity. In order to answer these questions, it is vital to understand the relationship between enzyme dose and bird performance.

IV. XYLANASE DOSE-RESPONSE

There is limited published information on xylanase dose responses, and if available, the information is often confined to two dose levels per experiment, which is clearly inadequate to establish an accurate dose response pattern. From recent studies, it is apparent that the xylanase response follows a nonlinear pattern, which can well be described by exponential functions (Figures 3 and 4).

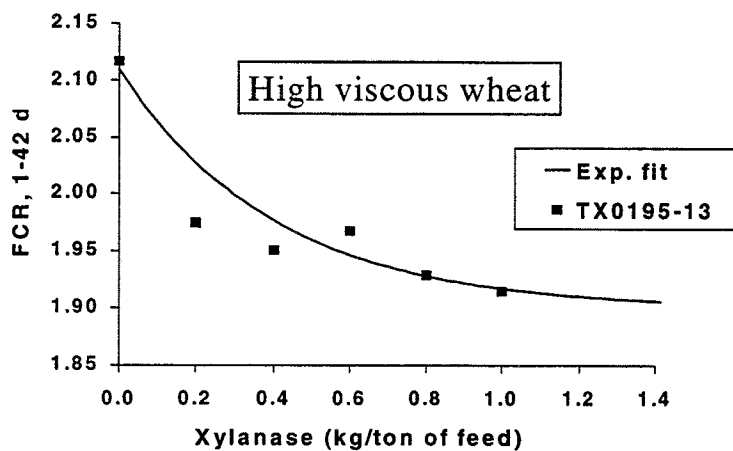


Figure 3. Feed:gain response in broilers to increasing xylanase dose (Chris Belyavin Technical, UK, broilers 1-42 days).

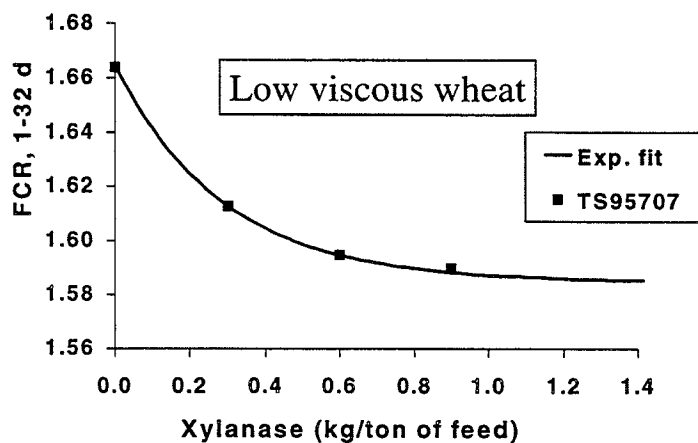


Figure 4. Feed:gain response in broilers to increasing xylanase dose (CNEVA Ploufragan, France, broilers 1-32 days).

It is noteworthy that a similar pattern was exhibited irrespective of the initial viscosity of the wheat. Hence the shape of the curve can be generalised for response prediction purposes. Differences in wheat quality as determined by extract viscosity determination merely result in an adjustment of the maximum response over the performance of the unsupplemented basal diet. Data are available only for the dose response to varying doses of Avizyme *Trichoderma longibrachiatum* xylanase. To date, it has not been established if different xylanases would follow a similar dose response pattern. Therefore one cannot generalise the present findings to cover other xylanase enzymes.

V. ECONOMIC DOSE OPTIMUM

On the basis of the finding outlined above, it is possible to accurately predict the response of a given wheat-based diet to xylanase addition. In the proposed model, (1) extract viscosity is determined as a means to identify the maximum response potential to xylanase, (2) adjustments to this response may be necessary to account for wheat inclusion rate, dietary fat source and processing conditions, and (3) the dose response prediction calculates the expected FCR depending on xylanase dose. This provides the basis for the calculation of the most economical enzyme dose, which is effectively done using the first derivative of the response curve along with prices for feed and enzyme. This gives a stepwise comparison of marginal income (through savings of feed cost with better FCR) *versus* marginal cost of the enzyme. The maximum profit is achieved where the marginal income from the respective last unit of enzyme added just matches the marginal cost of the enzyme. This approach, rather than using maximum performance as the parameter, gives the most relevant decision-making information for the commercial user of xylanase in wheat-based diets for broilers. It is meant to be used as a tool to optimise the benefit from using wheat as well as the xylanase in broiler feeds, and it can be employed to run various simulations on the effects of different wheat qualities or different feed and enzyme prices on the economic dose optimum.

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ENHANCED NUTRITIONAL VALUE OF SUNFLOWER MEAL FOR BROILERS FROM AN EXOGENOUS ENZYME PREPARATION

Y.G. LIU and C.A. ADAMS

Summary

On a worldwide basis, sunflowers rank as the second most important oilseed crop, being exceeded only by soybeans. Although sunflower is relatively new, it is rapidly gaining acceptance as a high-quality, plant protein feedstuff and competes with other oilseed meals. Meal from pre-pressed, solvent extracted, dehulled seeds contains about 440 g/kg of protein, as opposed to 280 g/kg for the whole seeds (Ensminger *et al.* 1990). Sunflower seed hulls are not easily removed, with the result that sunflower meal (SFM) is high in fibre, ranging from 150 to 240 g/kg. Traditionally, this meal is fed to ruminants. Recently, as the price of soybean meal has increased steadily, the introduction of sunflower meal into animal feeds, as a partial substitution for the expensive soybean meal, has become of great interest to many countries. Sunflower meal is a good protein source, but has a high fibre content which frequently restricts its use in feed for monogastric animals. Relatively low levels of up to 50 g/kg in broiler diets, 70 g/kg in layer diets and 100 g/kg in pig feeds have been used in the past. An extensive research programme has been conducted to develop a fibre-digesting enzyme system, Kemzyme HF, for broiler diets using SFM.

I. DEVELOPMENT OF A FIBRE-DIGESTING ENZYME SYSTEM

(a) In vitro studies with beet pulp materials

Fibrous components were prepared from beet pulp and feed-quality sunflower meal. These fibre fractions were then used as substrates to develop a suitable enzyme system for their degradation. Digestibility of the substrates was followed in a system where the solubility was measured. Dried beet pulp (200mg) was put into a glass tube and incubated in 20 mL of citrate buffer (pH4.6). Different enzyme preparations were added. The tubes were shaken and incubated at 40°C for 24 h. Afterwards the solutions were filtered and the filters were dried in an oven at 60°C. The weight of material remaining in the filter was the amount of undigested beet pulp. Results are presented in Table 1. In the absence of added enzymes only 2% of the material was solubilised, whilst cellulase enzyme alone or a commercial enzyme mixture gave some improvement in solubility to a maximum of 25%. The Kemzyme HF product, which is a complex mixture of cellulase and hemicellulase, solubilised about 90% of the beet pulp (Table 1).

It is possible that the particle size of the beet pulp might be important in this solubility test. Therefore, some of the material was finely ground prior to use in the solubility determination. As shown in Table 2, there was a slight effect of particle size at the lowest concentration of Kemzyme HF. At levels of 5-20 activity units there was an improved solubilisation of the beet pulp material compared to one activity unit, but grinding the beet pulp gave no advantage. The Kemzyme HF product achieved a high degree of solubilisation when the amount of substrates used varied by 15 fold from 200 to 3,000 mg (Table 3).

Table 1. Degree of solubilisation of dried beet pulp in the presence of different enzyme mixtures.

Treatment	Solubilised material (%)
Control	2.07
Cellulase A	15.41
Cellulase B	25.32
Commercial enzyme mixture	15.84
Kemzyme HF	90.09

Table 2. Degree of solubilisation (%) of ground or un-ground beet pulp in the presence of Kemzyme HF.

Enzyme activity	Type of beet pulp	
	Whole dried	Ground dried
1	55.4	69.0
5	86.5	82.7
10	89.9	88.0
20	90.1	88.4

The data in Tables 2 and 3 demonstrate that the Kemzyme HF is a powerful enzyme combination for fibre degradation. It is very effective even at low levels. This was also demonstrated by the sugar release test where 10g of substrate was incubated at 37°C for one hour. In this test the enzyme product was applied not only to beet pulp, but also to sunflower extract and grass pellets. After incubation the amount of reducing sugars was measured against a blank control. There was a linear increase in the release of reducing sugars with increasing levels of Kemzyme HF in all three substrates up to 2.6, 5.5 and 2.0 times the unsupplemented value in the mixtures supplemented with 10 activity units of Kemzyme HF in the beet pulp, sunflower extract and grass pellets respectively. The increased amounts of reducing sugars are considered to be derived from polysaccharides as a result of enzyme action. Therefore, not only is the fibre in the samples solubilised, but it is also broken down into simple sugars which should be available for nutritional purposes.

Table 3. Degree of solubilisation (%) of different amounts of beet pulp with a constant amount of Kemzyme HF.

Amount of beet pulp (mg)	Type of beet pulp	
	Whole dried	Ground dried
200	90.1	88.4
500	85.7	81.3
1000	83.8	81.7
2000	79.5	80.1
3000	71.5	77.1

(b) *In vitro* studies with fibre fractions extracted from sunflower meal

Further *in vitro* studies were conducted with neutral detergent fibre (NDF) and neutral detergent and acid detergent fibre (NDADF) fractions extracted from feed quality

sunflower meal. These fibre fractions were used as substrates for the Kemzyme HF. Both fibre fractions were attacked by the enzyme preparation, although the NDADF, which contains primarily the insoluble, indigestible cellulose fractions, was digested to a greater extent than the NDF fraction (10.0 cf 4.8%). The fibre-digesting enzyme system was designed to have its maximum effect upon the NDADF fraction of the sunflower meal.

II. BROILER TRIALS

In the first broiler trial, sunflower meal was supplemented with lysine, methionine and oil to make a feed ingredient called Napenerg. The Napenerg was then used to replace 60% of the soybean meal that was usually used in Hungarian broiler diets. The trial results are listed in Table 4. The diets with Napenerg and Kemzyme HF resulted in significantly better performance in terms of liveweight gain and feed conversion ratio (FCR) compared to the control diet with the normal amount of soybean meal ($P < 0.05$). Supplementing with the enzyme product increased the final weight by 8.3% (from 1841 to 1994 g) and improved FCR by 2.7% (from 2.24 to 2.18). The use of Napenerg without the addition of the enzyme product did not reduce final live weight but FCR was inferior compared to that obtained from the soybean meal control diet. This was likely due to the increased amount of fibre in the sunflower meal diet.

In a second broiler trial, a maize and soybean meal diet was used as the control. In the experimental diets, one third of the soybean meal was replaced by sunflower meal in the starter diet (0-28 d) and half replaced in the finisher diet (29-42 d). The diets were iso-energetic and iso-nitrogenous, but with different contents of crude fibre, varying from 32.7 to 45.4 g/kg in the starter diets and from 31.6 to 50.0 g/kg in the finisher diet. These differences in fibre content clearly affected broiler performance (Table 5). The sunflower meal replacement resulted in decreased weight gain and increased feed conversion ratio (FCR). However, when the diet was supplemented with Kemzyme HF there was a considerable improvement in broiler performance (Table 5). The daily growth rate was close to that obtained from the maize/soybean meal diet and the FCR was close to that of the standard diet.

Table 4. Effect of adding Kemzyme HF on the performance of broilers fed a diet in which sunflower meal replaced soybean meal.

	Live weight (g)	FCR
Soybean meal diet	1841 ^b	2.24 ^{ab}
Sunflower meal diet	1905 ^{ab}	2.48 ^a
SFM+Kemzyme HF	1994 ^a	2.18 ^b

Means within columns with different superscripts are significantly different ($P < 0.05$)

Table 5. Effect of adding Kemzyme HF on the performance of broilers fed on a diet using sunflower meal to replace 33% soybean meal in starter feed and 50% in finisher diet.

	Daily gain, g		0-42 d performance	
	0-28 d	29-42 d	ADG, g	FCR
Soybean meal	36.2 ^a	53.7 ^a	42.0 ^a	2.10 ^b
Sunflower meal	19.7 ^c	21.3 ^b	20.7 ^b	3.33 ^a
SFM+Kemzyme HF	32.3 ^b	51.9 ^a	38.3 ^a	2.23 ^b

Means within columns with different superscripts are significantly different ($P < 0.05$)

In the third broiler trial, the control feed was based on maize and soybean meal whilst the experimental feeds contained 200 g/kg sunflower meal (expeller cake) in both the starter and finisher feeds. Amino acid contents (methionine and lysine) were adjusted. This sunflower meal had 300 g/kg of protein, 150 g/kg of fibre and 90 g/kg of fat. A metabolism study was conducted to determine the excreta:feed (E:F) ratio from these feeds and the effect of enzyme supplementation. Supplementation of the sunflower diet reduced the E:F ratio from 45.2 to 38.1 ($P<0.05$), the latter value being essentially similar to that of the control diet (37.4).

In a performance trial with these diets (Table 6), whilst the decrease in liveweight with high levels of sunflower meal was not significantly improved by the addition of the enzyme preparation, the depression in feed efficiency on the sunflower based diet was considerably improved ($P<0.05$) by the inclusion of Kemzyme HF.

Table 6. Effect of Kemzyme HF and high levels of sunflower meal on broiler performance.

	Treatment		
	Control	Sunflower meal	SFM + Kemzyme HF
Live weight (g)	1720 ^a	1669 ^b	1686 ^{ab}
Feed conversion ratio	2.24 ^a	2.38 ^b	2.24 ^a

Means within a row with different superscripts are significantly different ($P<0.05$).

In the above broiler trials, substitution of soybean meal by sunflower meal decreased feed prices substantially.

III. CONCLUSION

Feed cost is a very important economic factor in poultry production. A fibre-digesting enzyme system, Kemzyme HF, has been developed which can partially degrade the fibrous components as demonstrated by studies with beet pulp and sunflower meal. Provided that amino acid (particularly lysine) requirements are met, this allows higher levels of sunflower meal to be incorporated into poultry diets. This study demonstrated that sunflower meal together with Kemzyme HF offers new possibilities to design more cost effective poultry feed formulations using alternative protein sources.

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HYDROLYSIS OF PLANT PHYTATE IMPROVES THE BIOAVAILABILITY OF NUTRIENTS IN POULTRY DIETS

L. CHRISTENSEN¹, G. HUYGHEBAERT² and D. PETTERSSON¹

Summary

Broiler chickens were fed diets with a reduced phosphorus content in a balance study in order to determine the effect of phytase supplementation on total tract digestibility of nutrients, such as Ca, P and amino acids. The results obtained indicate that by phytase supplementation of poultry diets it is possible to reduce the output of N and P to the environment by improved nutrient retention.

I. INTRODUCTION

In order to reduce the output of nutrients, such as nitrogen and phosphorus, to the environment a novel approach as regards monogastric feeding strategies and dietary formulations is needed. Evidently the first step would be to optimise diets based on digestible amino acids, thereby reducing the nitrogen surplus currently existing in most monogastric diets. Secondly, appropriate feed enzymes should be used in order to improve nutrient digestibility, thereby reducing faecal nutrient losses.

This paper reports results from a trial with phytase supplementation of a broiler diet in order to improve the digestibility and retention of macro nutrients.

II. PHYTATE

Phytate consists of a myo-inositol molecule, also referred to as phytic acid. This molecule may bind phosphorus but also other minerals. When bound to the phytic acid, minerals become unavailable. This was well documented for Ca, Cu, Fe, Mg, P and Zn from both animal and human experiments (Reddy *et al.*, 1982; Sandberg *et al.*, 1982).

The phytate phosphorus (pP) content may, on average, vary between 60% to 70% of the total phosphorus (tP) content in our commonly used cereals. However, the variation in pP content becomes higher when also considering the content in protein rich feedstuffs such as lupins or rape seed meal.

The digestibility of the non-phytate fraction, i.e. the potentially available phosphorus (aP), ranges between approximately 60% to 80% and even for mineral phosphorus supplements such as di-calcium phosphate the digestibility is not 100%, as sometimes is erroneously assumed in diet formulations. As a consequence of the low phosphorus digestibility a large portion of this important mineral is not retained in the body but instead excreted, together with other minerals that also may be bound to the phytate molecule. Investigations have shown that phytate in addition may impede protein digestibility, presumably by binding the protein molecule or by inactivating trypsin, and that phytate also possibly may inhibit α -amylase activity (Knuckles and Betschart, 1987; Mroz *et al.*, 1994).

¹ Novo Nordisk A/S, Novo Allé DK-2880 Bagsvæd, Denmark.

² Rijksstation voor Kleinveeteelt, Burg. V. Gansberghelaan 92, B-9820 Merelbeke, Belgium.

III. MATERIALS AND METHODS

Male broiler chickens were used in a balance trial. Groups of 4 birds (16 days old) were assigned randomly to the respective treatments with 5 replicates on each treatment. All birds were weighed and allocated to the respective pens (groups) in such a way that differences in group weights were minimised. A balance trial was carried out between days 22 to 26 according to the European reference method (Bourdillon *et al.*, 1990).

The broilers received 3 mash diets with increasing phosphorus content as control diets and the diet containing the lowest level of dietary phosphorus (Diet 1 at 4.1 g P/kg diet) was supplemented with 250, 500 or 750 FYT/kg to test the effect of phytase supplementation. All diets were fed at a level of 90% of *ad libitum* intake. Excreta were quantitatively collected on a daily basis.

A heat treated wheat (cultivar Tarnax) was used to minimise the effects of endogenous phytase, and soybean meal was the only other dietary fibre containing component (Table 1).

Table 1. Composition and nutrient content of the basal diets

	Diet 1 ¹	Diet 2	Diet 3
<i>Ingredient (g/kg)</i>			
Heated wheat	574.2	572.3	570.5
Soybean meal	308.3	308.3	308.3
Animal fat	79.6	79.6	79.6
CaCO ₃	13.4	10.1	6.9
Soybean oil	10.0	10.0	10.0
Di-calcium phosphate		5.0	10.0
NaCl	3.4	3.4	3.4
DL-Methionine	1.3	1.3	1.3
Vitamins and trace elements	10.0	10.0	10.0
<i>Nutrient content²</i>			
ME (MJ/kg)	13.10	12.95	13.00
Crude protein (%)	227	227	227
Phosphorus (%)	4.1	5.0	5.9
Calcium (%)	6.5	6.5	6.5

¹ Diet 1 was fed without phytase supplementation or with increasing levels of phytase (250, 500 or 750 FYT/kg diet).

² Determined content.

IV. RESULTS

Phytase supplementation generally resulted in an increased digestibility of both calcium and phosphorus (Table 2). The determined digestibility of the mineral phosphorus source used in this experiment was 69%.

In comparison with the unsupplemented control diets (Diet 1 and Diet 2) phytase supplementation with 500 FYT/kg diet significantly improved nitrogen retention. Although not statistically significant, higher apparent digestibility rates were observed for essential amino acids as a result of phytase supplementation at a level of 750 FTU/kg diet (Table 3).

Table 2. Digestibility (%) of calcium (Ca) and phosphorus (P) and nitrogen retention (g N/kg feed intake), in broiler chickens fed diets with different levels of phosphorus and phytase supplementation.

	Digestibility coefficients		Nitrogen retention
	Ca	P	
<i>Control diets</i>			
Diet 1 (4.1 g P/kg)	49 ^d	47 ^f	17.2 ^a
Diet 2 (5.0 g P/kg)	51 ^d	51 ^e	17.3 ^a
Diet 3 (5.9 g P/kg)	54 ^c	54 ^d	17.7 ^{abc}
<i>Phytase levels (FTU/kg diet)</i>			
Diet 1 (250)	56 ^c	57 ^c	17.3 ^{ab}
Diet 1 (500)	61 ^b	63 ^b	18.1 ^{bc}
Diet 1 (750)	66 ^a	65 ^a	18.2 ^c

^{abcdef} Means within a column with unlike superscripts differ significantly (P<0.05).

Table 3. Digestibility (%) of essential amino acids in broiler chickens fed Diet 1 with or without phytase supplementation at 750 FTU/kg.

Amino Acid	Diet 1 (4.1 g P/kg diet)	
	Unsupplemented	Phytase (750 FYT/kg)
Lysine	79.2	80.5
Threonine	74.7	75.8
Methionine	85.2	86.4
Cystine	70.7	71.8
Tryptophan	79.3	80.2
Isoleucine	80.9	82.4
Leucine	81.7	82.8
Phenylalanine	82.9	83.6
Valine	76.7	78.5

V. DISCUSSION

The presented results regarding Ca and P digestibility as well as nitrogen retention are in accordance with earlier findings. Improved amino acid digestibility rates have also been demonstrated in other trials with broiler chickens receiving phytase supplemented diets. In addition increased digestibility rates and retention of Cu, Fe and Zn on phytase supplementation have also been demonstrated in broilers (Windisch and Kirchgessner, 1996). There are also results available from rat experiments which indicate that phytase supplementation may reduce the accumulation of toxic metal ions such as cadmium (Cd) in the liver (Rimbach, *et al.*, 1995). The latter results need to be further corroborated in experiments with other animals. The indications that starch digestion may be improved as a result of phytase supplementation (Knuckles and Betschart, 1987), also needs to be investigated further in *in vivo* trials.

The current results indicate that phytase supplementation of broiler diets can significantly improve the digestibility and retention of nutrients such as Ca, P and N and that amino acid digestibility may also be improved.

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A REVIEW OF THE ANTINUTRITIONAL EFFECTS OF PHYTIC ACID ON PROTEIN UTILISATION BY BROILERS

A.K. KIES¹ and P.H. SELLE²

Summary

The antinutritional properties of phytic acid on protein utilisation in broilers and the capacity of microbial phytase to counter these effects are reviewed. The practical significance of these antinutritional effects has been shown in a series of studies in poultry. The relevance of protein-phytate complexes should be taken into account when dietary addition of phytase is considered.

I. INTRODUCTION

Phytic acid is a ubiquitous compound in vegetable feedstuffs where it serves as a phosphorus (P) depot. As monogastrics essentially lack endogenous phytase activity consequently phytate P is poorly digested and is excreted, increasing the P load on the environment. Microbial phytase (Natuphos®) was introduced in the Netherlands in 1992 as a feed enzyme to facilitate the reduction of P levels in effluents from intensive animal operations. However, recent data indicate that phytic acid also has a negative effect on protein utilisation and that this effect can be reduced by the dietary addition of phytase. Available data on these aspects are reviewed in this paper.

II. PHYTIC ACID - PROTEIN COMPLEX

The binding of protein by phytic acid to form protein-phytate complexes which reduces the solubility and digestibility of the complexed proteins has been recognised for some time. Champagne (1988) stated that at pH values lower than 5 (less than the isoelectric point of the protein), the anionic phosphate moieties of phytic acid bind strongly by electrostatic interactions to the cationic groups of protein to form insoluble complexes. The protein binding sites are believed to be provided by residues of lysine, histidine and arginine. At intermediate pH levels above 5, protein-phytate complexes probably are ternary structures where protein, mainly *via* histidine, is bound by mineral-phytate complexes involving calcium, magnesium and zinc. At pH values greater than 10 this ternary structure is disrupted; the protein is released and the mineral-phytate complex precipitates out.

It is probable that protein-phytate complexes exist inherently in vegetable feedstuffs to varying extents. Champagne (1988) cites evidence that phytic acid binds with proteins of soyabean, wheat, rapeseed and peanut but not with proteins of rice bran, maize germ and cottonseed meal. Cosgrove (1966) reported the extraction of a crystalline protein-phytic acid compound from beans with a P content of 4.8 g/kg which is substantially less than the P component of phytic acid (282 g/kg). This suggests that protein may be bound by phytic acid in a ratio of 50:1.

Recent *in vitro* data generated by Jongbloed *et al.* (1997) suggests that the *de novo* formation of protein-phytate complexes in the gut at pH values of 2-3 may be an important factor. At this pH soluble proteins are substantially precipitated out presumably by phytic

¹ Gist-brocades AgriIngredients Group, Delft, The Netherlands.

² BASF Animal Nutrition, Auburn, NSW, 2214.

acid forming complexes. Prior incubation of the feedstuff with phytase largely prevents this reaction. Incubation of the precipitated proteins with pepsin increased their solubility but this reaction was accelerated considerably by the addition of phytase. The implication is that phytase, in addition to releasing bound proteins, may reduce the formation of protein-phytate complexes within the gut by prior hydrolysis of phytic acid.

Considerable *in vitro* evidence exists which shows that phytic acid inhibits the activity of proteolytic enzymes. Caldwell (1992) reported the negative effects of phytic acid and calcium on the activation of trypsinogen and the stability of trypsin. The formation of protein-phytate complexes by phytic acid binding with endogenous enzymes may decrease the functionality of the enzymes and should be reduced or restored by phytase cleaving phytic acid.

Mroz *et al.* (1995) manipulated substrate levels in pig diets by substituting soyabean meal (3.9 g phytate P/kg) with rapeseed meal (7.0 g phytate P/kg). The resultant increase in dietary phytate P from 2.2 to 2.7 g/kg was associated with a trend towards reductions in both trypsin activity and nitrogen (N) digestibility. However, the main effect of the addition of a crude microbial preparation (800 PTU/kg) was to significantly ($P < 0.05$) increase both N digestibility (from 76.5 to 79.3%) and trypsin activity (from 4.51 to 5.00 units). Although yet to be confirmed, this work suggests that the body of *in vitro* data may have *in vivo* relevance.

The possibility that phytic acid may bind with supplemental amino acids to form "synthetic amino acid-phytate complexes" in practical diets was recently investigated *in vitro*. Rutherford *et al.* (1997) incubated lysine monohydrochloride with rice pollard as the source of phytic acid with and without added phytase. Incubation without phytase reduced the recovery of free lysine from 112 to 89 mg/100mL whereas with phytase addition the recovery was increased to 101 mg/100mL. Thus, the indication is that lysine was bound by compounds within rice pollard but partially released by phytase, which is consistent with the original hypothesis. Similar data with three amino acids (lysine, methionine and threonine) supplementing conventional diets has subsequently been generated by these workers (unpublished data).

III. EFFECTS OF PHYTASE ON PROTEIN UTILISATION

The hydrolysis of phytic acid by phytase releases phytate-bound proteins and may explain the "protein effect" of phytase. The first report of microbial phytase having a protein effect in poultry (van der Klis and Versteegh, 1991) was in laying hens. Levels of 250 and 300 FTU phytase/kg significantly ($P < 0.05$) increased the ileal absorption of nitrogen from 79.3 to 80.9%. It is well documented that phytase is very effective in improving overall P availability in layers. In this study, about 65% of dietary phytate-P was released by added phytase.

In broilers offered diets based on sorghum and soyabean meal, Farrell *et al.* (1992) showed that the dietary addition of microbial phytase (750 FTU/kg) resulted in a significant ($P < 0.05$) overall increase in N utilisation from 55.9 to 57.6% and an associated trend ($P < 0.10$) towards increased N retention from 56.0 to 57.0%. The protein contents of the diets were 188, 230 and 273 g/kg. The phytase response in N utilisation varied widely but corresponded to an average increase of 3.8 g/kg protein. It was suggested that the phytase response in N utilisation may have led to the increased feed intake by the broilers.

Kornegay (1996) investigated the effect of phytase on the ileal digestibility of amino acids in a factorial design with three levels of protein (170, 200 and 230 g/kg) and four levels of microbial phytase (0, 250, 500 and 750 FTU/kg). The broilers received diets based on

maize and soyabean meal containing 4.5 g/kg nonphytate P (nP) with a Ca : nP ratio of 2:1. Increasing protein levels linearly depressed the digestibility of all amino acids. The ileal digestibility of all essential amino acids, with the exception of methionine, were linearly increased ($P < 0.05$ to 0.001) by the addition of phytase. The average digestibility of methionine in the three control diets (without added phytase) was 94.6% and it was the most digestible essential amino acid.

In a broiler study, Ravindran and Bryden (1997) assessed the effects of phytase on the apparent ileal digestibility of nitrogen, nine essential amino acids and nitrogen retention. Three dietary substrate levels (2.9, 3.7 and 4.4 g/kg phytate P) were offered by increasing the inclusion rate of rice pollard, which contained 17 g/kg of phytate P, to 54 and 108 g/kg at the expense of wheat and sorghum. Increasing levels of phytate P or phytic acid significantly reduced N retention and the ileal digestibilities of N and all amino acids ($P < 0.03$ to < 0.001) with the exception of arginine ($P < 0.07$). This demonstrates the importance of dietary substrate levels and the negative effect phytic acid has on protein digestibility.

The addition of phytase (400 and 800 FTU/kg) significantly increased the digestibility of N and the amino acids ($P < 0.001$) and apparent N retention. However, this effect was more pronounced at the lower nP level of 2.3 g/kg than at the adequate level of 4.5 g/kg, resulting in significant interactions between phytase and the dietary nP level. The increase in nP was achieved by the dietary addition of approximately 12 g/kg dicalcium phosphate. Thus, it appears that inorganic Ca and/or P may have a negative, direct or indirect, impact on phytase activity. It is noteworthy that phytase permits the reduction of Ca and P levels in poultry diets.

The analysed crude protein level of the diets was 217 g/kg. Phytase increased the apparent ileal digestibility of N from 80.55 to 82.73% which corresponds to an increase of 4.73 g/kg of crude protein; in the lower nP diets the increase was from 80.50 to 83.05% which corresponds to 5.53 g/kg of N. Interestingly, the phytase responses, when expressed as apparent nitrogen retention (as % of intake), were numerically greater. Overall phytase increased protein retention from 53.1 to 56.2% which corresponds to 6.77 g/kg of N and in the lower nP diets the increase was from 54.9 to 58.3% or 7.36 g/kg of crude protein. There is a clear need to quantify the protein effect of phytase and the above differences suggest that protein deposition/retention data may be a more appropriate basis than ileal digestibility data to achieve this objective.

Biehl and Baker (1997) adopted an alternative approach by feeding broilers maize-based diets deficient in amino acids with either soyabean meal or peanut meal as the primary protein source. With the maize/soya diets, phytase (1200 FTU/kg) improved feed conversion ($P < 0.05$) of the amino acid-deficient diet but not of the adequate diet. It was concluded that phytase had a small, but significant, positive effect on the utilisation of methionine, threonine, lysine and/or valine from soya protein. Importantly, the results were similar irrespective of which amino acids were considered to be limiting. However, phytase failed to generate growth or conversion responses in the amino acid-deficient diets when peanut meal was the protein source. It may be relevant to note that phytic acid is more widely distributed throughout the seeds of soyabeans than peanuts. In this study the addition of phytase (1200 FTU/kg) to dehulled soyabean meal numerically increased true amino acid digestibility values for caecetomised roosters of ten amino acids by nearly 2 percentage units (mean: 90.4 to 92.2%) but the differences were not statistically significant.

Schutte *et al.* (1997) have generated positive amino acid data based on total tract digestibility values in broilers following the addition of phytase. Excreta amino acid digestibility data is routinely used in broiler formulations by the feedmilling industry in the Netherlands.

IV. CONCLUSIONS

Recent evidence indicates that phytic acid has a negative effect on the digestibility of proteins and amino acids in poultry. While the basis of this effect is not completely defined, the capacity of phytic acid to complex with proteins appears to be the core factor. Microbial phytase improves protein digestibility, probably by releasing bound proteins and/or reducing the extent to which they are complexed by phytic acid within the gut environment.

Two recent studies have demonstrated improvements in the ileal digestibility of amino acids in response to phytase supplementation. These responses were more evident in the Australian than in the American study which could be due to the fact that the American diets were inherently more highly digestible and would have contained lower levels of phytic acid than the Australian diets; these two factors may be related. In the Australian study, it is noteworthy that with the inadequate nP diets, which contained less inorganic Ca and P (essentially as dicalcium phosphate), phytase generated better responses than in the adequate nP diets. The adequate nP diets contained similar amounts of dicalcium phosphate and limestone to the American diets. The possible impact of inorganic Ca and P on the effectiveness of phytase requires further investigation but the enzyme does permit their reductions in the diet and narrow Ca:P ratios should be adopted with the addition of phytase.

The "protein effect" of Natuphos phytase needs to be quantified to facilitate its application in practical diets. This effect is likely to be more pronounced in diets of moderate, rather than superior, protein quality and in diets with relatively high levels of phytic acid or phytate-P (>3.0g phytate-P/kg). Clearly these variables must be taken into account in any estimation of protein replacement values for phytase in poultry diets.

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FEEDING BEHAVIOUR OF 10 WEEK OLD PULLETS FOLLOWING BEAK TRIMMING AT HATCH

P. C. GLATZ¹, C. A. LUNAM² AND J. L. BARNETT³

Summary

This experiment examined whether chronic pain in the beak was still evident in 10 week-old layer pullets beak trimmed at hatch by comparing their feeding and pecking ability with a control group not trimmed. There was no evidence to indicate the beak was sore as beak trimmed pullets pecked more at the cage ($P < 0.05$) and had more toe pecks and preening bouts ($P > 0.05$). Beak trimmed pullets also made more feed pecks and feed pecks per gram ($P > 0.05$), suggesting beak trimmed pullets had reduced mechano reception than control pullets. While these data suggest no reduction in any pecking behaviours and thus an absence of chronic pain it is not known if the force applied when pecking differed between treatments.

I. INTRODUCTION

Beak trimming is performed early in the life of commercial hens to decrease injuries caused by the behavioural vices of cannibalism, bullying, and feather and vent pecking. It involves partial removal of the upper and lower beak using an electrically heated blade. Objections to the use of beak trimming include its removal of sensory receptors, with a subsequent reduction in feed intake (Glatz and Lunam, 1994) and pecking efficiency (Gentle *et al.*, 1982); permanent loss of temperature and touch responses (Gentle, 1986b) and behavioural evidence (hyperalgesia and guarding behaviour) for persistent pain (Duncan *et al.*, 1989; Gentle *et al.*, 1990). A major concern is that beak trimming may induce chronic pain. Traumatic-neuromas in the beak stump after trimming have been implicated as a cause of chronic pain in commercial hens (Beward and Gentle, 1985; Gentle, 1986a). In our studies (Lunam *et al.*, 1996) neuromas were present in all beaks at 10 weeks, but neuromas were not found at 70 weeks after moderate trimming at hatch. As neuromas were not observed in adult hens that had been moderately trimmed at hatch, our results indicate that they develop and persist for at least 10 weeks, before resolving. This experiment was undertaken to determine if there was behavioural evidence for the presence or absence of persistent neuromas by making studies on feed and pecking behaviours of pullets 10 weeks after trimming at hatch.

II. MATERIALS AND METHODS

(a) Beak trimming and imprinting

Twenty day-old chickens were beak trimmed soon after hatch according to Australian Model Code of Practice for the Welfare of Domestic Poultry (1995). A heated blade on a commercial electric beak trimming machine cut and cauterised for half the upper beak and one third of the lower beak for two seconds. Twenty control chickens were not beak trimmed.

¹ Pig and Poultry Production Institute, Roseworthy Campus, Univ of Adelaide, SA 5371.

² Flinders University of South Australia, Bedford Park, SA 5042.

³ Victorian Institute of Animal Science, Werribee, Victoria 3030.

Chickens were housed in a battery brooder and imprinted for 60 mins daily to peck at a red plastic block (lxbxd= 3 x 3 x 3 cm) in the first 5 days of life. Red block was held by handler and moved around in brooder to encourage chickens to follow and peck at the moving block. At the end the daily imprinting period chickens approached and pecked at the block when it was placed on the brooder floor. It was hypothesised that if beaks were sore 10 weeks after trimming, birds would peck less at red block than control chickens. At 4 weeks of age pullets were transferred into rearing cages. Birds were fed a chick starter mash from 0-4 weeks and a pullet grower mash thereafter.

(b) Feeding behaviour

Twenty birds (10 weeks of age) from each treatment were deprived of feed 1 hour prior to testing. Individual birds were placed in a test cage with a feed hopper attached. Pullets had been previously placed in the test cages for three 30 min training periods prior to the test with feed available. Feed was weighed into a hopper and pecks at feed and billing of feed (bird flicking feed from side to side in hopper) was monitored for 30 minutes with a video recorder. In addition bouts of dust bathing and number of head shakes were recorded and feed pecks per gram of feed consumed calculated. A higher number of head shakes was interpreted as increased sensitivity to pain (Gentle *et al.*, 1990). Filming of birds took place from 900-1800 h and took 2.5 days to complete.

(c) Pecking behaviour

The same birds used for the feeding behaviour studies were used for the pecking studies. Twenty birds (10 weeks of age) from each treatment were deprived of feed 1 hour prior to testing. Individual birds were placed in a test cage with a feed hopper attached. Pullets had been previously placed in the test cages for three 30 min training periods prior to the test with feed available but without the presence of the red block. Red block was placed in bottom of a feed hopper with no feed provided. Pecking at red block, pecking at feed hopper, toe pecking, pecking at cage and preening bouts were monitored for 30 min with a video recorder. Filming of birds took place from 900-1800 h and took 2.5 days to complete.

(d) Data analyses

The experiment was analysed to determine the effect of beak trimming on behaviour of pullets using the general linear models procedure using SAS GLM (Statistical Analysis Systems Institute Inc., 1988).

III. RESULTS AND DISCUSSION

If pullets were suffering from chronic pain, they would be expected to engage in less pecking and preening bouts. There was no evidence for this in the present experiment. Beak trimmed birds pecked more at the cage ($P < 0.05$) and, while the differences were not statistically significant, also pecked more at the feed hopper (Table 1) and had more toe pecks and preening bouts ($P > 0.05$). In addition they also made more feed pecks and feed pecks per g (Table 2) of feed consumed from hopper ($P > 0.05$). Beak trimmed and control chickens showed little interest in pecking at the red block. While these data suggest no reduction in any pecking behaviours and thus an absence of chronic pain, it is not known if the force applied when pecking differed between the treatments. Beak trimming may alter the sensory

perception of the bird (Gentle *et al.*, 1982) and reduce the ability of the pullet to pick up food (Workman and Rogers, 1990). We observed a decrease in pecking efficiency in our experiment with beak trimmed pullets making both more pecks at the food ($P>0.05$) and more pecks per gram of food consumed ($P>0.05$) than the control pullets (Table 2). Contrary to a previous report (Gentle *et al.*, 1990) there was an increase in pecks ($P<0.05$) made at the cage and the feed hopper ($P>0.05$) by the beak trimmed pullets compared to the controls despite neuromas being observed in trimmed beaks at 14 weeks (Lunam *et al.*, 1998). It should be borne in mind, however, that Gentle *et al.* 1990 made their behavioural assessment of chronic pain 6 weeks after beak trimming. Our studies were made 10 weeks after trimming when it is likely that neuromas were in the process of resolving (Lunam *et al.*, 1996).

The increased pecking and visual stimulation gained by pullets as a result of feeding appeared to encourage more dust bathing ($P>0.05$) in beak trimmed pullets compared to controls (Table 1). Petherick *et al.* (1993) report the sight of a dusty substrate is an important factor in initiating dust bathing. Feather pecking is more likely to occur when birds are dust bathing (Vestergaard *et al.*, 1993), presenting a problem for the egg farmer. Beak trimmed pullets are more likely to dust bathe and be pecked than controls, yet if beak trimming is not undertaken cannibalism will occur.

Our study has shown that beak trimmed birds in the test cage had significantly fewer head shakes than the controls. This indicates that trimmed birds are less fearful than untrimmed ones, supporting the findings of Lee and Craig (1991).

These results suggest that 10 weeks after beak trimming pullets may not be suffering the degree of chronic pain originally thought despite presence of neuromas. Instead our results indicate that beak trimmed pullets have reduced mechano reception ability as a result of the altered beak shape.

Table 1. Pecking behaviour in a 30 minute period (feed unavailable) for 10 week old pullets beak trimmed at hatch versus control group not trimmed. P= probability in one-way analysis of variance.

Treatment	Cage pecks	Hopper pecks	Toe pecks	Preening bouts	Pecks at red block
Control	10.5a	2.5	0.3	10.0	1.5
Beak trim	16.1b	6.4	0.7	12.8	1.1
P	0.02	0.09	0.22	0.59	0.70

Table 2. Feeding behaviour in a 30 minute period following feed withdrawal for 10 week old pullets beak trimmed at hatch versus control group not trimmed. P= probability in one-way analysis of variance.

Treatment	Feed pecks per g	Feed intake (g/bird)	Feed pecks	Feed bills	Dust bathes	Head shakes
Control	98	3.3	281	88	8	2.8a
Beak trim	139	3.1	368	88	17	1.1b
P	0.09	0.70	0.23	0.99	0.12	0.01

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RESPONSES OF MEAT CHICKENS TO DIETS DIFFERING IN TOTAL AND DIGESTIBLE THREONINE CONTENT

P.F. MANNION, R. PEREZ-MALDONADO and D.J. FARRELL

Summary

Formulating diets using digestible amino acids (DAA) in lieu of total amino acids (TAA) is gaining wide acceptance. The anticipated benefits include providing amino acids closer to the bird's true requirement and a more accurate selection of foodstuffs based on their amino acid contribution. A comparison of the two systems was undertaken in modelling the responses of male and female broiler chickens from 3 to 24 days of age to diets differing in threonine concentration.

Amino acid digestibility of each ingredient was measured and subsequently two series each of five diets based on TAA or DAA were prepared using a summit diet and a low protein dilution mixture in each case.

The significant responses in liveweight gain, food intake and food conversion ratio (FCR) to the diets in each series were attributed to differences in the intake of total and digestible threonine. The DAA series of diets gave a better mean FCR than the TAA series, confirming the expected benefit of increased efficiency. Curves describing the responses to both total and digestible threonine were calculated and estimates of threonine requirement for maximum liveweight gain and minimum FCR are given.

I. INTRODUCTION

Based on current estimates of amino acid requirements, threonine is often the next limiting essential amino acid after methionine and lysine in typical Australian diets for broiler chickens. The requirement for threonine is often most cheaply met by its direct addition to the diet but the current cost of manufactured threonine is considerably greater than that of either lysine or methionine.

There are no published data comparing responses of broilers to intakes of total and digestible threonine from diets based solely on Australian foodstuffs and from which estimates of requirements can be made.

II. MATERIALS AND METHODS

(a) Chickens and management

Broiler chickens obtained from a commercial hatchery (Steggles Ltd) which had been vent sexed and vaccinated for IB were reared in wire cages in a heated room from day-old. At three days of age chickens within each sex were individually weighed and assigned to groups, each with a weight range of 5 g. The heaviest and lightest were discarded and the remainder were randomly assembled into groups of eight chickens, such that each group had a liveweight of similar mean and variance. Each group was housed in a wire cage until the completion of the experiment at 24 days of age.

Queensland Poultry Research and Development Centre, Alexandra Hills, Qld 4161

Food and water were available *ad libitum*. Illumination was for 23 h and hot-air brooders provided heating and ventilation. The temperature of the experimental rooms was regulated thermostatically in accordance with the comfort of the birds (30 to 23°C). Food intake and liveweight were measured after 21 days on the experimental diets. Birds which died were individually weighed and the food residue recorded at that time in the affected cage.

(b) Experimental diets

There were 12 experimental diets comprising two series each of six diets. The two series were based on formulations using TAA and DAA contents of the dietary ingredients. Within each series a range in threonine contents was obtained by blending a summit diet with a low protein dilution mixture in appropriate proportions to give five diets. The summit diet (Diet 1) of each series had a threonine content that was 1.2 times the requirement estimated for maximum growth; the other essential amino acids were no less than 1.4 times requirement. The sixth diet in each series was produced by adding sufficient synthetic L-threonine to the diet with least threonine (Diet 5) to raise its threonine content to that of the next highest diet in the series. The dilution mixture was formulated to contain the same calcium, phosphorus and calculated metabolizable energy content as the summit diets. The compositions of the two summit diets and of the dilution mixture are given in Table 1.

Table 1. The ingredient composition (g/kg) of the summit diets and dilution mixture.

	Summit*	Summit*	Dilution
	TAA	DAA	
Sorghum	351.9	438.9	-
Soybean meal	354.0	418.7	-
Cottonseed meal	100.0	16.7	-
Meat and bone meal	16.4	77.3	-
Fishmeal	28.3	-	-
Faba bean	58.7	-	-
Ground rice husk	-	-	268.9
Starch	-	-	200.0
Sucrose	-	-	434.3
Soybean oil	44.9	22.8	50.0
Limestone	11.3	5.7	10.9
Dicalcium phosphate	9.8	-	24.9
Sodium chloride	3.4	3.0	5.0
Ferrous sulphate	1.6	-	-
DL-methionine	5.1	4.3	-
L-lysine HCl	5.7	4.6	-
L-isoleucine	1.0	0.5	-
L-leucine	1.3	-	-
L-histidine	-	0.5	-
L-valine	0.6	1.0	-
Mineral premix	1.5	1.5	1.5
Vitamin premix	4.5	4.5	4.5

* Summit diet formulated from total amino acid values (TAA) or digestible amino acid values (DAA).

(c) Experimental design

There were 96 experimental units enabling the 12 dietary treatments, applied to two sexes, to be randomly allocated within four positional replicates in a randomized block layout.

(d) Amino acid digestibility

Amino acid digestibility was measured on each feed ingredient using ISABrown caecectomized cockerels in a latin square design with five birds per feed ingredient (Green and Kiener, 1989). Details have been published (Anonymous, 1989).

III. RESULTS AND DISCUSSION

Liveweight gain, food intake and FCR measured over the 21 d experimental period and combined across sexes all responded significantly to the different dietary levels of both total and digestible threonine. Interactions with sex were not significant ($P>0.05$).

The fact that these observations represent responses to the level of dietary threonine in both the total and digestible series is supported by the significant responses ($P<0.05$) for liveweight gain and food intake to added L-threonine in diet 6 compared with performance on diet 5 in each series. Although the threonine content of diet 6 was the same as that of diet 4 in each diet series, performance was not the same because of restrictions imposed by the second most limiting amino acid.

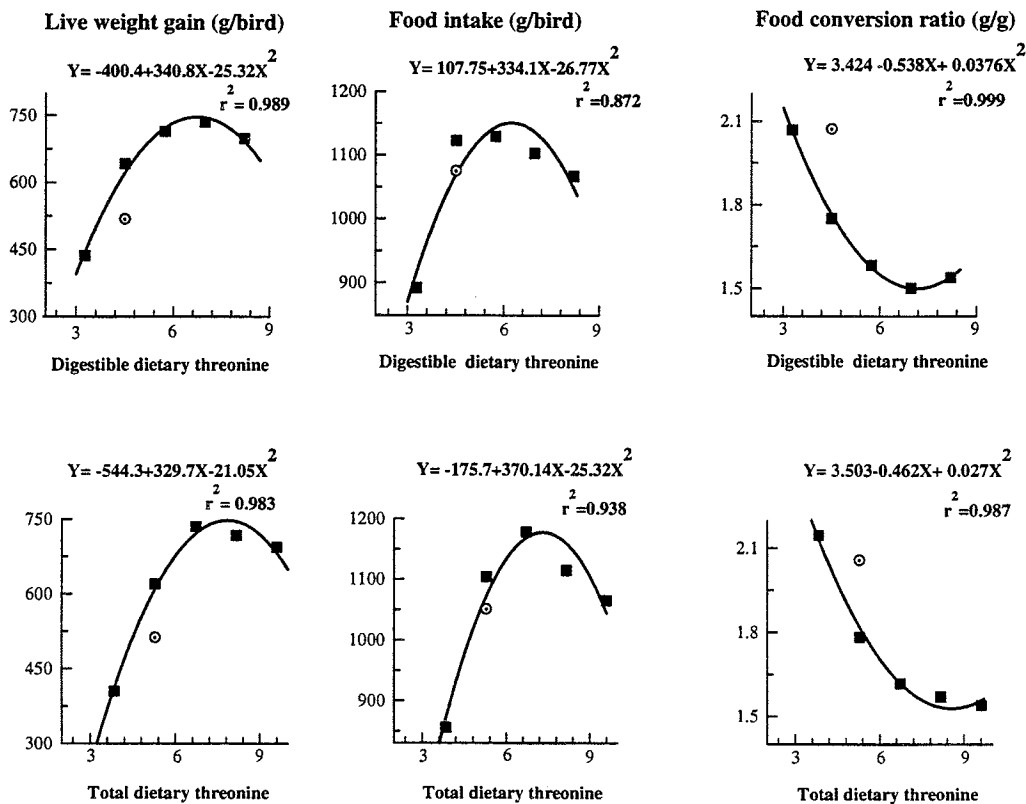


Figure 1. Fitted response curves to digestible and total dietary threonine (g/kg).
○ Response of birds fed Diet 6.

Diets formulated using DAA gave a significantly ($P<0.05$) better mean FCR than for the TAA series although there were no differences ($P>0.05$) in liveweight gain or feed intake. None of the interactions were significant ($P>0.05$), indicating that the superior FCR for the DAA series applies over the range of dietary threonine concentration and to both male and female chicks. A quadratic model accounted for at least 98% of the variation between diets for both liveweight gain and FCR and at least 87% for food intake. The regression equations and r^2 values are given

in Figure 1. Neither the linear nor quadratic coefficients of the regression equations differed ($P>0.05$) between the diet series for any of the parameters shown in Figure 1. Maximum liveweight gain and minimum FCR values together with their corresponding dietary threonine concentration were calculated (Table 2). These values can be compared with total threonine requirements in the range 6.3 to 7.7g/kg reported by Kidd and Kerr (1996).

Table 2. Calculated maximum liveweight gain (g/bird), minimum FCR (g/g) and corresponding dietary threonine concentration (g/kg). Data combined across sexes.

	Liveweight gain	Dietary threonine	FCR	Dietary threonine
TAA series	747.3	7.8	1.528	8.6
DAA series	746.5	6.7	1.499	7.2

To better compare the responses to both total and digestible threonine, liveweight gain and FCR were regressed on the daily intake of digestible threonine calculated for each diet in both series (Figure 2). At the time of writing a full statistical comparison of these regressions had not been undertaken. However, they appear to confirm the results from the analysis of variance that formulating diets on the basis of digestible amino acids improved FCR while having no effect on liveweight gain.

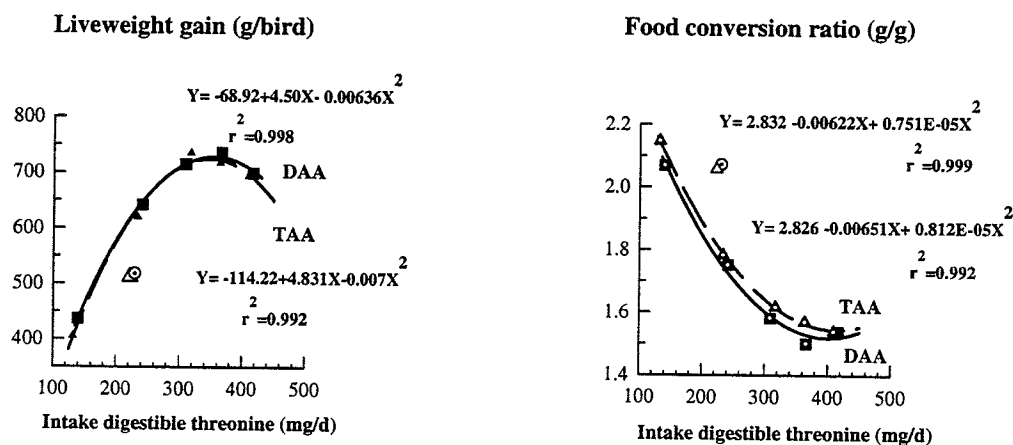


Figure 2. Fitted response curves for both DAA and TAA series to intake of digestible threonine. \circ , Δ Response of birds fed Diet 6 of DAA or TAA series, respectively.

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NUTRITIVE VALUE OF LUPINS FOR BROILERS

R.J. HUGHES¹, A. KOCHER¹ and M. CHOCT²Summary

The nutritive value of Australian lupins for broiler chickens was assessed in a series of classical apparent metabolisable energy (AME) studies each of 7-days duration. AME values (MJ/kg dry matter) for lupin kernels ranged from 12-15 for *L. albus* cv Kiev mutant, 8-11 for *L. angustifolius* cv Gungurru, and 6.5-10.5 for *L. angustifolius* cv Danja. The AME for wholeseed of *L. luteus* was 11.4 MJ/kg dry matter. Large differences due to season and locality were evident, which is common in cereals and legumes. Removal of the seed coat (de-hulling) increased AME by approximately 24 and 15% in white and sweet lupin, respectively. The seed coats of these species do not contain anti-nutritive factors but did have an energy dilution effect. Inclusion of wholeseed of Gungurru lupin at 200 g/kg in a wheat-based diet and at 300 g/kg in a maize-based diet did not have deleterious effects on growth, feed efficiency or digesta viscosity. Excreta moisture was significantly increased by inclusion of wholeseed of Gungurru lupin at 200 g/kg in wheat-based and maize-based diets.

I. INTRODUCTION

Early cultivars of lupin were high in alkaloids and bitter components, and regarded as unsuitable as feedstuffs for monogastric animals. Genetic selection of varieties low in these anti-nutritive factors has led to the widespread use of lupins as a protein source for poultry (van Barneveld and Hughes 1994). Earlier studies by Karunajeewa and Bartlett (1985) and Brenes *et al.* (1993) showed that broiler chickens can tolerate up to 250 g/kg of low-alkaloid lupin-seed meal without adversely affecting growth, provided there are adequate supplements of lysine and methionine. Conversely, Johnson and Eason (1991) warned that inclusion of *L. angustifolius* in diets at 150 g/kg at the expense of soybean meal could reduce growth and feed efficiency. Constraints are often placed on the maximum inclusion level of lupins in broiler diets due to the incidence of wet excreta which can pose a health risk to the birds through respiratory stress from high ammonia and potential carcass down-grading due to breast blisters and hock burns.

A series of classical energy balance studies were conducted to determine the effects of (a) different seasonal conditions and growth sites of various species and cultivars of lupin, (b) removal of the seed coat, (c) dietary inclusion rate of lupin and NSP, and (d) addition of feed enzyme products on dietary AME, growth performance, viscosity of ileal digesta and excreta moisture of commercial broiler chickens given diets containing up to 300 g/kg lupin.

II. MATERIALS AND METHODS

Day-old mixed sexed broiler chickens were raised in floor pens on a commercial starter crumble to 21 days and then on finisher pellets. At 24 days of age, birds were weighed in groups of five and transferred to metabolism cages in controlled temperature rooms for classical AME studies involving quantitative measurements of feed intake and excreta output.

¹ SARDI, Pig and Poultry Production Institute, Nutrition Research Laboratory, University of Adelaide, Roseworthy, SA 5371.

² Department of Animal Science, University of New England, Armidale, NSW 2351.

Semi-purified diets containing cereal grains (wheat, sorghum or maize), casein, and minerals and vitamins were fed for seven days. In each study, dietary treatments were replicated six times. The first three days enabled the chickens to adapt to the cages and the feeds. During the following four days, all excreta were collected and dried. Birds were weighed in groups at the end of the seven day period. Feed intake was measured during the adaptation and collection phases of the study. Ileal digesta were centrifuged at 12,000 g for 30 minutes. Viscosity was determined on thawed supernatant using a Brookfield DVIII viscometer at 25°C with a shear rate 5-500 s⁻¹. Dry matter (DM) contents of samples of pelleted and milled feeds were measured. Gross energy values of dried excreta and milled feeds were measured with a Parr isoperibol bomb calorimeter

III. RESULTS AND DISCUSSION

Large variation in AME was observed between different species and cultivars grown in different localities and under different growing conditions (Table 1). De-hulling increased AME by approximately 24 and 15% in Kiev and Gungurru, respectively. Differences between species were due partly to the higher oil content of Kiev (93 g/kg) compared with Gungurru (53 g/kg), and partly to the higher total non-starch polysaccharide (NSP) content of Gungurru (550 g/kg) compared with Kiev (390 g/kg) (van Barneveld, pers. comm.).

Table 1. Apparent metabolisable energy (AME MJ/kg dry matter) of lupin species and cultivars grown at different sites in Western Australia and in different years.

Year	Species	Cultivar	Form	No. of samples	Mean	Lowest value	Highest value
93	<i>L. albus</i>	Kiev †	Kernel	1	12.4	-	-
	<i>L. angustifolius</i>	Gungurru	Kernel	4	9.0	8.2	11.0
	<i>L. angustifolius</i>	Danja	Kernel	3	8.4	6.5	10.5
94	<i>L. albus</i>	Kiev	Kernel	3	14.8	14.6	14.9
	<i>L. angustifolius</i>	Gungurru	Kernel	3	10.8	9.8	12.3
95	<i>L. albus</i>	Kiev	Wholeseed	1	10.7	-	-
	<i>L. albus</i>	Kiev	Kernel	1	12.6	-	-
	<i>L. angustifolius</i>	Gungurru	Wholeseed	1	6.3	-	-
	<i>L. angustifolius</i>	Gungurru	Kernel	1	8.3	-	-
96	<i>L. luteus</i> ‡	Unknown	Wholeseed	1	11.4	-	-

† Grown in New South Wales

‡ Grown under irrigation.

Samples of Kiev and Gungurru lupin grown on three farms in WA in 1994 (see Table 1) were put through an experimental separator to obtain relatively pure samples of seed coat. The hull material was added to a sorghum-based semi-purified diet at 70g/kg (equivalent to 300-350 g/kg whole lupin). The reductions in AME and growth performance (Table 2) and lack of effect on viscosity of ileal digesta and excreta moisture indicate that seed coats of Kiev and Gungurru were low in energy but had no discernible anti-nutritive effects.

Deleterious effects of lupin NSP on dietary AME, growth performance, ileal viscosity and excreta condition are clearly evident in the results shown in Table 3. The lupin NSP levels are equivalent to addition of wholeseed Gungurru at 100-300 g/kg. Feed enzyme product (Avizyme 1300, 1 kg/tonne) reduced the moisture content of excreta when lupin NSP was low (50 g/kg) but not at higher levels. A particularly interesting aspect of these results is

the large increase in ileal viscosity due to enzyme action in the presence of high dietary levels of lupin NSP.

Table 2. Effects of dietary inclusion of seed coat from samples of *L. albus* cv Kiev mutant and *L. angustifolius* cv Gungurru grown on three farms in WA on feed conversion ratio (FCR, g feed/g gain), growth rate (GR, g/bird 24-31 days), dietary AME (MJ/kg dry matter), ileal viscosity (IV, cP) and excreta moisture (EM, g/kg). Means (n = 6) having a common superscript are not significantly different (P<0.05).

Farm	Species	FCR	GR	AME	IV	EM
Dongara	<i>L. albus</i>	2.25	328	14.2 ^{bc}	4.0	605
	<i>L. angustifolius</i>	2.29	323	14.3 ^{bc}	4.3	615
Walkaway	<i>L. albus</i>	2.26	338	14.3 ^{bc}	4.4	625
	<i>L. angustifolius</i>	2.27	330	14.2 ^c	4.5	592
Balla	<i>L. albus</i>	2.39	312	14.2 ^{bc}	4.5	619
	<i>L. angustifolius</i>	2.21	342	14.4 ^b	5.1	602
Sorghum/casein basal diet		2.21	318	15.2 ^a	2.8	635

Table 3. Effects of dietary inclusion rate of NSP (0, 50, 100 and 150 g/kg) extracted from *L. angustifolius* and addition of enzyme product on feed conversion ratio (FCR, g feed/g gain), growth rate (GR, g/bird 24-31 days), dietary AME (MJ/kg dry matter), ileal viscosity (IV, cP) and excreta moisture (EM, g/kg). Means (n = 6) having a common superscript are not significantly different (P<0.05).

NSP	Enzyme	FCR	GR	AME	IV	EM
0	No	2.19 ^a	316 ^{ab}	14.5 ^a	4.6 ^a	593 ^a
0	Yes	2.14 ^a	329 ^a	14.7 ^a	2.9 ^a	608 ^a
5	No	2.34 ^{ab}	316 ^{ab}	12.8 ^b	4.6 ^a	685 ^c
5	Yes	2.27 ^a	331 ^a	13.1 ^b	6.8 ^a	666 ^b
10	No	2.78 ^c	268 ^c	11.0 ^{cd}	8.0 ^a	727 ^d
10	Yes	2.65 ^{bc}	275 ^{bc}	11.5 ^c	27.0 ^b	725 ^d
15	No	3.15 ^d	219 ^d	10.3 ^e	12.3 ^a	762 ^e
15	Yes	3.24 ^d	217 ^d	10.5 ^{de}	37.2 ^b	766 ^e

Increasing levels of *L. angustifolius* cv Gungurru in both wheat and maize-based diets resulted in decreased dietary AME, and increased excreta moisture (Table 4). Similar effects were observed in sorghum-based diets (Hughes, unpublished data). In addition, the wheat-based diets with 300 g/kg lupin had significantly poorer feed conversion and growth, with an associated increase in ileal viscosity. In a similar study with *L. angustifolius* cv Gungurru at 300 g/kg in a maize-based diet, a commercial enzyme product (Ronozyme W, 400 g/tonne) improved dietary AME by 0.4 MJ/kg dry matter and raised ileal viscosity from 7.4 to 14.6 cP. All of the lift in energy was attributed to action of enzyme on the maize component. When a combination of enzymes was used (Ronozyme W and Ronozyme VP, each at 200 g/tonne), dietary AME and ileal viscosity were not significantly different from the control diet.

Several other experiments with various commercial feed enzyme products failed to yield consistent results in terms of improvement in AME, growth, or feed efficiency in short-term metabolism studies. Perhaps longer feeding periods with larger numbers of chickens are necessary to detect small but significant differences from both statistical and commercial

view-points. In several studies with enzyme products, large increases in viscosity of ileal digesta indicate that lupin NSP substrates were partially depolymerised by enzymes but not broken down to simple sugars able to be absorbed by the gut. Development of enzyme technology is required for cost-effective treatment of anti-nutritive effects of lupin NSP.

Table 4. Effects of cereal and dietary inclusion rate of *L. angustifolius* on feed conversion ratio (FCR, g feed/g gain), growth rate (GR, g/bird 24-31 days), dietary AME (MJ/kg dry matter), ileal viscosity (IV, cP) and excreta moisture (EM, g/kg). Means (n = 6) having a common superscript are not significantly different (P<0.05).

Cereal	Lupin	FCR	GR	AME	IV	EM
Maize	0	2.12 ^{ab}	328 ^a	15.3 ^a	2.9 ^a	623 ^a
	10	2.02 ^a	358 ^a	14.2 ^b	4.5 ^{ab}	648 ^{ab}
	20	2.09 ^a	359 ^a	13.0 ^c	6.3 ^{abc}	695 ^{cd}
	30	2.20 ^{ab}	335 ^a	12.0 ^{de}	4.2 ^{ab}	726 ^{de}
Wheat	0	1.98 ^a	327 ^a	12.3 ^d	8.4 ^{bc}	668 ^{bc}
	10	2.01 ^a	316 ^a	12.5 ^d	10.4 ^{cd}	700 ^{cd}
	20	2.14 ^{ab}	329 ^a	11.7 ^{ef}	11.4 ^{cd}	713 ^{de}
	30	2.33 ^b	266 ^b	11.4 ^f	14.3 ^d	737 ^e

IV. CONCLUSIONS

The results of these short-term metabolism studies indicate that Australian sweet and white lupins are valuable sources of protein and energy for inclusion in broiler diets. The seed coats of these species do not contain anti-nutritive factors but did have an energy dilution effect. Wholeseed of Gungurru lupin can be included up to 200 g/kg in wheat-based diets and up to 300 g/kg in maize-based diets without detriment to growth performance of chickens. Inclusion of 200 g/kg wholeseed of Gungurru lupin in either wheat- or maize-based diets has the potential to promote wet droppings. Enzyme technology for degrading lupin NSP needs development.

V. ACKNOWLEDGMENTS

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PRACTICAL LEVELS OF INCLUSIONS OF FOUR GRAIN LEGUMES IN BROILER DIETS IN A LARGE-SCALE EXPERIMENT

R.A. PEREZ-MALDONADO, P.F. MANNION and D.J. FARRELL

Summary

An experiment was undertaken on a semi-commercial scale to identify practical inclusions of field peas, faba beans, chick peas and sweet lupins in steam-pelleted broiler starter and finisher diets. Faba beans (100-180 g/kg) and sweet lupins (120 g/kg) were generally superior to field peas (200-300 g/kg) and chick peas (100-220 g/kg) in growth rate and feed conversion ratio (FCR). Sweet lupins (120 g/kg) did not respond to a feed enzyme addition when cold pelleted. Viscosity of digesta and visual scoring of pen litter at 42 days did not indicate unduly wet excreta or high gut viscosity. Recommended upper practical levels (g/kg) of the grain legumes for broilers in both the starter and finisher diets were: field peas 300, chick peas 100, faba beans 200 and sweet lupins 100.

I. INTRODUCTION

There is a need to strengthen and expand the feed base for the Australian poultry industries in view of recent forecasts (see Farrell, 1997) and the escalating prices of protein concentrates.

We have reported at this meeting (Perez-Maldonado *et al.*, 1997a) and at another meeting (Perez-Maldonado *et al.*, 1997b) experiments using diets containing four different grain legumes at various levels in broiler starter and finisher diets. Stepwise inclusions ranged from 120 to 360 g/kg. Field peas and faba beans gave significantly higher mean growth rates and lower mean feed conversion ratios (FCR) than chick peas and lupins. There was a significant effect of level of inclusion of some grain legumes in starter and finisher diets. For sweet lupins and chick peas the highest inclusion level gave depressed performance in starter diets and for chick peas in the finisher diets. Viscosity measurements of intestinal digesta supernatant indicated that sweet lupins, even at the lowest level of inclusion (120 g/kg), were unsatisfactory because they caused wet droppings.

On the basis of these results, a large-scale experiment was designed in which the same grain legume seeds as those used previously were included in broiler starter and finisher diets at two levels. The first was that used in commercial practice in Australia; the second was the upper limit of inclusion that was considered safe to be used by industry.

II. MATERIALS AND METHODS

Four thousand hatch-day broiler chickens of a commercial strain were used and were sexed on site. The experiment comprised of eight treatments each with four replications that were allocated in blocks of four to 64 pens (1.5 x 5.0 m) with 60 healthy chicks per pen and the sexes reared separately. An electric bar heater provided warmth and wood shavings (7 cm depth) were used as litter. Dead and culled chicks were replaced during the first three days.

Queensland Poultry Research and Development Centre, Alexandra Hills, Qld 4161.

At 21 days, 16 broilers per treatment were killed and viscosity measurements were made on digesta supernatant in the small intestine of birds on the sweet lupin diets only (Perez-Maldonado *et al.*, 1997a). The four grain legumes were: field peas (cv. glenroy), chick peas (cv. amethyst), sweet lupins (cv. gungurru) and faba beans (cv. fiord). Their inclusion levels and composition of the diets are given in Table 1.

Feed was available *ad libitum* from three tube feeders and water from six dry-cup waterers per pen. Diets were steam-pelleted and crumbled to 21 days when feed consumption and bodyweight were recorded. Steam-pelleted finisher diets were fed to 42 days of age when feed consumption and body weight were recorded. One diet containing sweet lupins plus a feed enzyme was cold pelleted. Diets were formulated with similar levels of energy and crude protein using the Feedmania computer program. Lighting was varied according to commercial practice in order to minimise mortality.

Table 1. Major ingredient composition of diets; minerals and vitamins were included and free amino acids where appropriate.

Inclusion	Starter diet							
	Field peas		Chick peas		Faba beans		Sweet lupins	
	20% ¹	30% ²	10% ¹	17% ²	10% ¹	18% ²	12% ¹	12% ^{2,3}
Ingredient								
Sorghum (CP 11%)	384	291	446	406	436	380	411	411
Wheat (CP 13%)	120	120	120	120	120	120	120	120
Meat and bone meal (CP 50%)	65	66	64	66	68	63	64	64
Soybean meal (CP 45%)	181	158	240	205	242	204	229	229
Grain legume	200	300	100	170	100	180	120	120
Inclusion	Finisher diet							
	Field peas		Chick peas		Faba beans		Sweet lupins	
	20% ¹	30% ²	15% ¹	22% ²	15% ¹	18% ²	12% ¹	12% ^{2,3}
Ingredient								
Sorghum (CP 11%)	480	406	415	370	400	378	402	402
Wheat (CP 13%)			120	120	120	120	120	120
Fish meal (CP 65%)			19	15	25	25	25	25
Meat and bone meal (CP 50%)	65	56	55	59	50	50	51	51
Soybean meal (CP45%)	90		130	100	70	50	30	30
Soybean meal (full-fat)	121	189	69	74	142	152	209	209
Grain legume	200	300	150	220	150	180	120	120

¹ Commercial practice; ² Maximum recommended; ³ Cold pelleted + feed enzyme.

III. RESULTS AND DISCUSSION

There was a significant ($P < 0.001$) effect of diet and sex of bird for all parameters shown in Table 2. However there was no diet x sex interaction ($P < 0.05$). Bodyweight at both ages was significantly higher ($P < 0.05$) on the diet containing 180 g faba beans/kg and bodyweight at both ages was lowest on the diet containing 180 g chick peas/kg. FCR was also lower on the 18% faba bean based diet. Sweet lupins showed no improvement in bird performance as a result of enzyme addition. Field peas, normally used as a control in grain legume diets, did not give expected results. FCR at 200 g/kg inclusion was the highest of all diets and growth rate was lower than for sweet lupins and faba beans. Carré *et al.* (1991)

showed that starch digestibility in field peas was influenced by cultivar and processing and the tannin content of this cultivar of field peas was found to be high (Perez-Maldonado *et al.*, 1997b). Tannins in faba beans have also been cited as the antinutritional factor responsible for poor chicken performance; recent work would indicate that these may not be the reason (Wareham *et al.*, 1993). Faba beans contain significant amounts of antinutritional factors, particularly vicine and convicine (Pettersson and Mackintosh, 1994) but these did not appear to have an adverse effect on performance.

Relative viscosity of digesta from the small intestine of birds on the two sweet lupin diets was the same (11.1, 10.8) but substantially less than found in previous experiments. Visual scoring (1-3) of litter in pens gave values of 1.25 without, and 0.75 with the feed enzyme. Perez-Maldonado *et al.* (1997b) found in broiler chickens aged 42 d that gut viscosity supernatant was 20.4 when sweet lupins were included in diets at 120 g/kg.

Table 2. Bodyweight, feed intake and feed conversion ratio (FCR) of broiler chickens (data combined for sexes) at 21 and 42 days (d) of age on starter (S) and finisher (F) diets with four grain legumes at two levels of inclusion.

Diets	S	F	Bodyweight (g)		Feed intake (g)		FCR	
			21d	42d	21d	42d	21d	42d
FP	20 ¹	-20	695 ^{ab2}	2081 ^{cd}	948 ^c	4120 ^a	1.471 ^c	2.040 ^a
FP	30	-30	698 ^{ab}	2088 ^c	1008 ^a	4090 ^{ab}	1.555 ^b	2.008 ^{ab}
CP	10	-17	672 ^{cd}	2085 ^{cd}	947 ^c	4000 ^{abc}	1.519 ^b	1.968 ^{bc}
CP	15	-22	656 ^d	2042 ^d	944 ^c	4024 ^{abc}	1.555 ^b	2.026 ^{ab}
FB	10	-18	684 ^{cb}	2133 ^b	966 ^{cb}	3970 ^{bc}	1.521 ^b	1.911 ^{cd}
FB	15	-18	711 ^a	2180 ^a	971 ^{cb}	4011 ^{abc}	1.469 ^c	1.888 ^d
SL	12	-12	678 ^c	2115 ^{cb}	981 ^{ba}	3951 ^c	1.560 ^{ab}	1.920 ^{cd}
SL	12E	-12E ³	674 ^{cd}	2120 ^{cb}	996 ^{ba}	3926 ^c	1.600 ^a	1.908 ^{cd}
LSD (P=0.05)			18.1	44.1	32.0	129.0	0.0425	0.0628

¹ FP = field peas %; CP = chick peas %; FB = field beans %; SL = sweet lupins %; ² Values with different superscripts are significantly different (P<0.05); ³ With feed enzymes.

Perez-Maldonado *et al.* (1997b) observed no difference (P>0.05) between groups of broiler chickens on finisher diets with these same grain legumes when data were combined for all levels of inclusion (120-360 g/kg). Similar comparisons at 21 days showed field peas and faba beans to be superior to sweet lupins and chick peas (Perez-Maldonado *et al.*, 1997a). However, in both previous small-scale experiments there was sometimes variation between inclusion rates within a grain legume.

Because starting weights were different between treatments at the commencement of the 21-42 d finisher period, the actual liveweight gains have been calculated and are given in Table 3. The results show much more clearly that field peas gave significantly (P<0.05) reduced weight gain compared with faba beans and sweet lupins.

The results of this experiment suggest that inclusion of sweet lupins at 120 g/kg in this experiment did not increase substantially gut viscosity nor was wet excreta observed as in our previous experiments. Pen litter was not unduly moist at the end of this experiment. This level might then be considered to be the maximum inclusion for this reason in finisher diets. The upper practical inclusion of faba beans is at least 180 g/kg and likely higher. Chick peas should not be included in practical diets at levels much above 100 g/kg and in our previous

experiments this amount caused significant pancreas enlargement. This is probably due to their high levels of trypsin and chymotrypsin inhibitors (Batterham *et al.*, 1993). Field peas may not give maximum bodyweight and FCR compared to faba beans at 200-300 g/kg even though field peas are not known to have any antinutritional factors.

Table 3. Bodyweight gain (21-42 d of broilers on the four grain legumes at two levels of inclusion.

Grain legume	Inclusion (g/kg)	Bodyweight gain (g)
Field peas	200	1385 ^{cl}
Field peas	300	1390 ^c
Chick peas	150	1412 ^{cb}
Chick peas	220	1385 ^c
Faba beans	150	1448 ^{ab}
Faba beans	180	1469 ^a
Sweet lupins	120	1437 ^{ab}
Sweet lupins ²	120	1446 ^{ab}

¹ Values with different superscripts are significantly different ($P < 0.05$).; ² With feed enzyme.

In summary, the recommended upper levels of inclusion (g/kg) for these cultivars of grain legumes in practical, steam-pelleted diets for broilers during the starter and finisher phases are: field peas 300, chick peas 100, faba beans 200 and sweet lupins 100. It is known that in practice wet droppings have been observed at this inclusion of sweet lupins and it might be prudent to reduce this in broiler starter diets. Finally, small-scale experiments may not give sufficiently precise information that can be transferred to the poultry industry without first undertaking experiments on the scale described here.

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FREE CHOICE FEEDING AND AMINO ACID REQUIREMENTS IN BROILERS

B. TJIPTOHARDJONO and R.A.E. PYM

Summary

Two experiments were conducted with a commercial broiler strain to determine responses in growth and body composition to free choice feeding and to dietary regimens in which amino acid allowances decreased at different rates from one-day-old to 40d using a summit-dilution technique. The two diets were isoenergetic (13.0 MJ ME/kg) but differed in crude protein (CP) level by a factor of two (240 and 120g CP/kg). In both experiments the efficiency of lean tissue growth rate in the free choice fed birds was as good or better than the blended diet groups. The free choice fed birds demonstrated a capacity to modify their intake of protein to meet altered requirements induced by changes in management.

I. INTRODUCTION

Chickens have been shown to possess a capacity for balancing their nutrient intake to reasonably meet their requirements when presented with a range of alternative feedstuffs containing all the essential nutrients (Rose and Kyriazakis, 1991). Practicalities dictate that the number of choices is limited, and the typical form that free choice feeding in poultry has taken is to offer the birds high-energy low-protein and low-energy high-protein components (e.g. Mastika and Cumming, 1987).

One of the potential uses of free choice feeding in broilers is as a means of meeting growth-related changes in nutrient requirements as the birds age. With the refinement of growth models and increasing information on nutrient requirements, a suggested approach to this is to offer the birds the choice between complete diets of similar energy content, but varying in amino acid concentration. In this way the bird has the opportunity to eat only as much protein as it needs as it grows from hatching to slaughter weight. This paper reports on two experiments in which measures were made of responses in growth and body composition to typical slaughter age in a commercial broiler strain, either allowed free access to, or given mixtures of, two isoenergetic diets varying in amino acid concentration.

II. EXPERIMENTAL PROCEDURE

In both experiments two diets were prepared, one a summit (one-day-old) diet with 240g CP, 16.8g lysine (total) and 13.0 MJ ME/kg and the other a 120g CP, 6.0g lysine and 13.0 MJ ME/kg dilution diet. The ingredient and nutrient composition of the two diets are given in Table 1. Based on information gained earlier in a broiler growth model strain characterisation study (Gous *et al.*, 1996), the summit diet was formulated to meet the nutrient requirements of day-old chicks of the strain used, and the dilution diet was formulated to have an appropriate amino acid balance for the strain.

In experiment 1, 200 one-day-old commercial male broiler chicks were allocated at random to 24 flat-deck cages in a hot-air brooder room at the Queensland Poultry Research and Development Centre, Alexandra Hills. The birds were given one of three dietary regimens in which the diet was changed every five days, by blending appropriate amounts of the summit and dilution diets together, to follow a linear decrease in dietary protein

concentration from 240g/kg at day-old to either 180 (H), 160 (M) or 140 (L) g/kg at 40 days, or a choice of the summit and dilution diet in a trough divided into two compartments. There were six replicate pens of eight birds of each sex per treatment to 20d of age.

Table 1 Ingredient (g/kg) and calculated nutrient (g/kg) composition of the summit and dilution diets used in the two experiments.

Ingredient	Experiment 1		Experiment 2	
	Summit	Dilution	Summit	Dilution
Sorghum	432.4	-	470.8	22.1
Wheat	250.0	62.1	150.0	51.7
Maize	-	842.0	-	825.1
Soyabean meal	163.7	6.7	225.4	9.3
Meat meal	54.7	67.2	50.7	69.3
Fish meal	70.0	4.3	75.0	-
Sunflower oil	19.5	-	20.4	-
Limestone	1.5	6.2	1.9	6.0
DL-Methionine	1.93	0.08	1.60	0.13
Lysine mono HCL	6.20	2.03	4.19	2.14
Salt	0.14	2.76	-	2.78
ME (MJ/kg)	13.0	13.0	13.0	13.0
Crude protein	240	120	240	120
Lysine (tot)	16.8	6.0	16.8	6.0
Methionine	5.71	2.10	5.61	2.10
Isoleucine	9.25	3.70	10.00	3.70
Threonine	9.80	4.10	9.04	4.10
Tryptophan	2.41	1.03	2.56	1.03
Linoleic acid	20.0	16.7	20.0	16.7
Calcium	10.0	10.0	10.0	10.0
Phosphorus (av)	6.0	4.5	6.0	4.5

At 20d of age the birds were transferred to 120 single cages in a fan-ventilated, temperature-controlled room at the Veterinary Science Farm, Pinjarra Hills. Here they continued to receive the diets within the respective regimens with dietary change every five days, with approximately 15 individual replicates per treatment. For the free-choice (FC) birds, the position of the two diets in the divided trough was altered every five days. Measures were made every five days from day-old to 40d of food consumption and body weights. The birds were killed at 41d for measurement of breast meat yield and abdominal fat pad weight. Data on weight gain, FCR, breast yield and abdominal fat were analysed using the Least Squares General Linear Model procedure of SAS.

The procedures adopted in experiment 2 were essentially similar, with the following exceptions: dietary changes were made every 7d; the 42d target dietary protein levels were 130 (L), 160 (M) or 190 (H) g/kg; the birds were killed at 43d for measurement of breast meat yield and abdominal fat pad weight; and the maize used in the dilution diets was roller- rather than hammer-milled as in experiment 1.

III. RESULTS AND DISCUSSION.

The effect of the four dietary regimens on growth rate (g/d) in the males in both experiments is shown in Fig. 1. A notable feature of the growth response in experiment 1 was a depression in growth rate during the 20-25d interval, greatest in the L group and least in the FC group. This is likely due to a delay in the birds' acclimatisation to the single cages.

The effect was much less apparent in the second experiment, although there was a marked depression in growth rate over the 35-42d interval in this experiment.

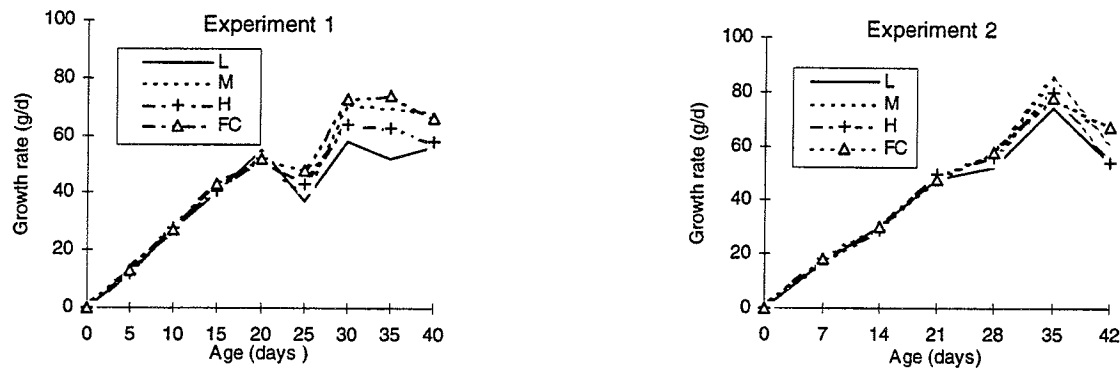


Figure 1 Growth rate (g/d) in the H,M,L and FC groups from 0-40 and 0-42d in the two experiments

The growth response in experiment 1 can be explained, in part at least, by protein consumption as shown in Fig. 2. In all groups the linear response in protein intake to 20d was interrupted on transfer to the single cages with a marked reduction in the L group, a plateauing in the M and H groups and a slight deviation from the initial rate of increase in the FC group. This growth and protein intake effect on transfer to the single cages was not evident in experiment 2.

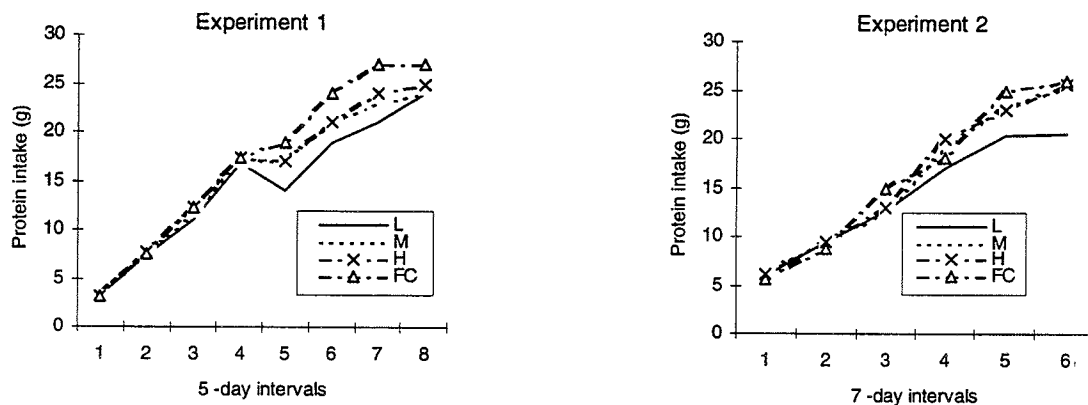


Figure 2 Protein intake (g/d) in the L,M,H and FC groups from 0-40 and 0-42d in the two experiments

Responses in growth rate, feed efficiency and body composition during the latter growth phase in both experiments are shown in Table 2. Growth performance from 20 to 40d in both experiments was greatest in the FC and M groups and least in the L groups, with the H groups intermediate. Feed conversion efficiency was also best in the FC birds in both experiments, with some between-experiment variation in the relative performance of the birds given the blended diet treatments. Breast yield was highest and abdominal fat lowest in the FC and H groups in experiment 1 and in the H group in experiment 2 ($P < 0.05$). In both experiments, the L group had the least breast yield and the greatest amount of abdominal fat ($P < 0.05$).

The relative consumption of summit and dilution diets by the free choice fed birds in the two experiments is shown in Fig. 3. In experiment 1 the birds ate more than twice as much summit as dilution diet, whereas in experiment 2, the consumption of the two diets was

Table 2 The effects of the four dietary regimens in the two experiments on 20-40d (Exp.1) and 21-42d (Exp.2) weight gain (g) and FCR, and 41d (Exp.1) and 43d (Exp.2) breast meat yield (g/kg) and abdominal fat (g/kg).

Experiment	Treatment	Weight gain	FCR	Breast yield	abdominal fat
1	L-140	1015 ^c	2.064 ^b	124.0 ^c	11.8 ^a
	M-160	1238 ^{ab}	1.946 ^a	132.8 ^b	9.1 ^b
	H-180	1139 ^b	2.080 ^b	139.1 ^a	7.1 ^{bc}
	Free choice	1306 ^a	1.887 ^a	141.3 ^a	6.0 ^c
	LSD _{0.05}	118	0.115	8.5	2.3
2	L-130	1259 ^b	1.983 ^c	190.3 ^b	14.4 ^a
	M-160	1425 ^a	1.831 ^b	193.8 ^{ab}	11.6 ^b
	H-190	1337 ^{ab}	1.768 ^{ab}	199.9 ^a	7.0 ^c
	Free choice	1420 ^a	1.707 ^a	193.9 ^{ab}	10.8 ^b
	LSD _{0.05}	95	0.087	7.6	2.8

Means within columns and experiments with different superscripts are significantly different ($P < 0.05$)

almost identical. Given the similarity in the nutrient and ingredient composition of the two diets in the two experiments (Table 1), it is likely that the physical form of the dilution diet in particular, due to the hammer- vs roller-milling of the maize, may have influenced the relative consumption of this diet in the two experiments. Palatability factors may have been involved.

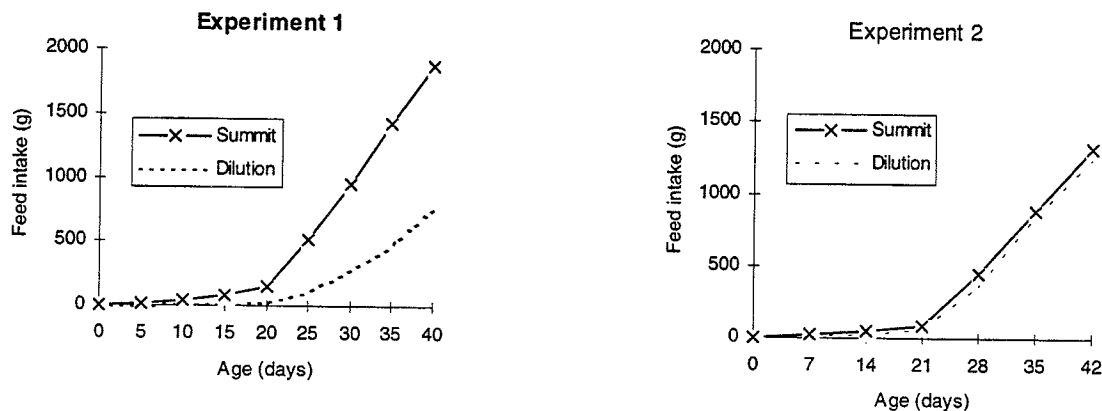


Figure 3. Relative intake (g/d) of summit and dilution diets in the two experiments

The improved lean tissue growth rate of the FC group in experiment 1 appears to be a reflection of their greater capacity than the blended diet groups, to maintain an adequate intake of protein on transfer to the single cage environment. The FC groups in both experiments performed as well or better than the blended-diet groups, suggesting that birds can regulate their intake of protein to achieve efficient rapid lean growth, and that given such choice, there is the opportunity for the birds to change their intake of protein to meet alterations to requirements for amino acids imposed by changes in external factors.

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IMPROVING THE QUALITY OF LOW VALUE LEGUMES BY AUTOCLAVING

K.G. WIRYAWAN and J.G. DINGLE

Summary

Protein quality and true metabolizable energy (TME) are lower in black gram and lupin compared with other legumes. An experiment to study the effect of autoclaving time on the nutritional quality of black gram and lupin was conducted. Male broiler chickens of 7 to 21 d old were fed isoenergetic and isoprotein diets which included black gram or lupin autoclaved at 125°C for 0, 5, 10, 15 or 20 min. Responses of black gram and lupin to autoclaving were different. Black gram was not affected by autoclaving. Net protein ratio (NPR) value increased by 10% and TME value by 6.5% when lupin meal was autoclaved for 5 min. Other methods should be pursued to improve the nutritive value of black gram.

I. INTRODUCTION

Apart from methionine deficiency, the presence of greater quantities of antinutritional factors (ANF) might be the cause of low NPR (Wiryawan and Dingle, 1995) and low TME values (Wiryawan and Dingle, 1996) of black gram (*Phaseolus mungo*) and lupin (*Lupinus angustifolius*) compared with other legumes.

Thermal inactivation of antinutritional factors in grain legumes has been studied. Methods such as autoclaving, toasting, and extrusion cooking improve the nutritive value of grain legumes. Studies with chickens showed that autoclaving smooth peas (*Pisum sativum*) for 3 min at 130°C and 170 kpa increased the apparent metabolizable energy (AME) value, and protein and starch digestibility (Conan and Carre, 1989). The weight gain of chickens fed a diet containing 75% autoclaved faba bean or field peas increased by 8 or 4%, and the feed conversion ratio (FCR) decreased by 10 or 11% compared with those fed raw bean or peas (Ernest, 1984). The effects of autoclaving, however, depend on both temperature and duration of heating. Excessive heating may reduce the availability of some amino acids, especially lysine. Van Barneveld (1993) reported that 26 to 40% of the lysine of peas was not available after heating at 135 and 150°C respectively. Therefore it is important to define the optimum heat treatment to maximize the inactivation of ANF and minimize the loss of amino acids.

The aim of this experiment was to study the effect of autoclaving on the quality of black gram and lupin for growing chickens.

II. MATERIALS AND METHODS

Black gram and lupin meals were spread to a depth of approximately 1.5 cm on stainless steel pans and heated to 125°C at 180 kPa for 0, 5, 10, 15 and 20 min in a standard laboratory autoclave and used as the sole sources of protein in isoenergetic diets containing nominally 10% crude protein. In protein quality tests, diets containing 100g/kg protein are usually applied, since more consistent and differential results have been obtained (Bressani, 1977). The respective dietary treatments (g/kg) were: BO, B50, B100, B150 and B200 for black gram and LO, L50, L100, L1500 and L20 for lupin. Dietary fibre content was adjusted with solka floc (Table 1).

Department of Animal Production, The University of Queensland, Gatton College, Qld 4345.

One-week old male meat chickens were randomly allocated into individual cages (one bird per cage, and eight chickens per treatment) in a temperature controlled room ($30\pm 1^{\circ}\text{C}$) and given unlimited access to diet and water for 14 days. Eight chickens were fed an isoenergetic protein-free diet as a negative control group. All chickens were weighed at 7 days and 21 days after an overnight fast. The responses to dietary treatments were assessed in terms of feed intake (FI), net weight gain (NWG) and NPR value. The NPR was calculated as (weight gain of the chickens fed the test diet + weight loss of the chickens fed protein-free diet) divided by protein intake (Bender and Doell, 1957).

Table 1. Ingredient composition (g/ kg) of the two diets.

Ingredients	Black gram	Lupin
Black gram	400.2	-
Lupin	-	319.0
Starch	446.1	559.2
Sunflower oil	73.1	41.0
Mineral and vitamin premix	2.0	2.0
Solka floc	39.1	36.0
Di-calcium phosphate	22.3	24.5
Limestone	16.7	16.5
DL-Methionine	0.6	1.7

Upon conclusion of the 14 days feeding trial, the chickens were randomly reallocated in the cages and fed a commercial diet for 3 days then left without feed for 24 h to empty their alimentary canals. True metabolizable energy (TME) value and apparent digestibility of protein (APD) of the diets were determined according to the methods of Sibbald (1979) with slight modification. The modification involved feeding a mixture of one portion of diet with one portion of water using a plastic syringe. It required only about 10-15 seconds to fill the crop with the feed mixture. A plastic tray was placed under each cage and the time recorded. The precise amount of test material consumed was the difference between the weight of the chicken before and after feeding. Each test material was fed to eight chickens. The excreta were collected 24 h after feeding. The excreta of two birds were pooled, freeze dried, left overnight at room temperature, weighed and ground to pass a 0.8 mm screen and kept for analysis of energy and nitrogen contents.

Nitrogen (N) content was analysed in an automatic nitrogen analyser using the combustion method (Sweeney, 1989). The faecal N was calculated as total N minus uric acid N. Crude protein (CP) content was calculated as $\text{N} \times 6.25$ and uric acid was determined according to the method of Marquardt (1983). Energy of feed and faeces was determined using a bomb calorimeter with benzoic acid as standard and correcting for total acid production by titration with 0.0709 N sodium carbonate solution. All samples were analysed in duplicate. The TME value and APD for each group of birds were calculated according to equation 1 and equation 2 respectively:

$$\text{TME (MJ / kgDM)} = \frac{\text{EI} - \text{FE} + \text{EEL}}{\text{FI}} \quad (1)$$

$$\text{APD (\%)} = \frac{\text{PI} - \text{PF}}{\text{PI}} \times 100 \quad (2)$$

where EI = energy intake, FE = faecal energy, FI = feed intake and EEL = endogenous energy loss of unfed birds, PI = protein intake, and PF = faecal protein.

Data were subjected to analysis of variance using the General Linear Model (GLM) and the Regression Procedures of the Statistical Analysis System (SAS) Institute, Inc. (1990) program.

III. RESULTS AND DISCUSSION

The FI, NWG, NPR, TME and APD are presented in Table 2. Significant interactions between legumes and autoclaving time on FI and NWG were observed. This suggests that the chickens' responses to each legume autoclaved for different durations were different. Although the NWG of chickens fed B5 diet was significantly greater ($P < 0.05$) than those fed B0 and B20 diets, overall NWG, FI and NPR values were not significantly affected by autoclaving time. On the other hand, an 18% increase in FI and a 50% increase in NWG were observed when the chickens were given the L5 diet compared to the L0 diet.

Table 2. Effect of autoclaving time (min) on FI (g), NWG (g), NPR, TME (MJ/kg) and APD (%) of black gram and lupin diets in young chickens.

Legume	Time	Dietary Treatments	FI	NWG	NPR	TME	APD
Black gram	0	B0	211.08 ^{bcde}	40.84 ^{cd}	3.75 ^{ab}	14.80 ^{ab}	76.72 ^a
	5	B5	265.46 ^{ab}	69.56 ^{ab}	4.04 ^{ab}	14.90 ^{ab}	72.23 ^a
	10	B10	231.48 ^{bcd}	57.05 ^{bc}	4.13 ^{ab}	14.51 ^{ab}	79.04 ^a
	15	B15	229.56 ^{bcd}	51.59 ^{bc}	3.82 ^{ab}	14.04 ^{bc}	76.03 ^a
	20	B20	208.43 ^{bcde}	41.59 ^{cd}	3.67 ^{ab}	13.56 ^c	69.28 ^{ab}
Lupin	0	L0	249.83 ^{abc}	57.66 ^{bc}	3.80 ^{ab}	14.23 ^{bc}	72.89 ^a
	5	L5	294.51 ^a	86.43 ^a	4.17 ^a	15.15 ^a	70.21 ^{ab}
	10	L10	196.45 ^{cde}	36.55 ^{cd}	3.79 ^{ab}	14.31 ^{abc}	68.71 ^{ab}
	15	L15	174.75 ^{de}	25.94 ^{de}	3.56 ^b	14.36 ^{abc}	70.39 ^{ab}
	20	L20	155.24 ^e	8.10 ^c	2.71 ^c	14.01 ^{bc}	60.97 ^b
SEM ¹	-		17.47	7.62	0.17	0.27	3.21

Means within columns with a similar superscript are not significantly different ($P > 0.05$)

¹ Standard error of the mean.

Regression analysis showed that the effect of autoclaving time on NPR of lupin meal was curvilinear fitting the equation: $NPR = 3.86 + 0.05x - 0.005x^2$ ($P < 0.001$; $R^2 = 0.49$), where x = autoclaving time (min), but the regression of autoclaving time of black gram on NPR was not significant. Nevertheless the maximum response on both regression curves occurred after 5 min heating but the response decreased with longer heating time. Yanez *et al.* (1986) reported a 51% and 29% reduction of available lysine and sulfur containing amino acids respectively after roasting (80 - 90°C) lupin meals for 10 to 40 min. Furthermore, it was reported that 26 to 40% of the lysine of peas was not available after heating at 135 to 150°C respectively (Van Barneveld *et al.* 1993).

Autoclaving did not significantly improve the TME value of black gram diets, but the TME value of L5 was significantly ($P < 0.05$) greater than that of the L0 diet. This is in agreement with a previous report (Boldadji *et al.* 1986). In line with Yanez *et al.* (1986), the APD was not improved by autoclaving. In fact, prolonged heating reduced ($P < 0.05$) the APD

value of both legumes. Products of the Maillard reaction might have contributed to a higher nitrogen level in the faeces.

There is no clear reason to explain the lack of positive response of black gram meal to autoclaving. It is possible that there were no heat-labile anti-nutritional factors in the sample of black gram meal. Black gram has been reported to be devoid of lectins (Reddy and Salunkhe, 1981). Although the black gram contained a relatively higher level of trypsin inhibitor than other legumes (Wiryanan and Dingle, 1996), this inhibitor may be resistant to heat treatment. An intensive study conducted by Padhye and Salunkhe (1980) showed that only 9% of the activity was lost when the trypsin inhibitor in black gram was heated at 100°C for 5 min.

In conclusion, the nutritive value of lupin can be improved by autoclaving for 5 min but other methods should be sought to improve the nutritive value of black gram.

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ADVANTAGE OF USING DIGESTIBLE PROTEIN AND DIGESTIBLE AMINO ACIDS TO FORMULATE POULTRY DIETS

M.A. ALI¹, A. KHATUN² and J.G. DINGLE¹

Summary

Azolla meal prepared from the aquatic fern azolla was included in layer diets at a level of 0, 50, 100, 150 or 200 g/ kg by replacing sesame meal on a total protein and total amino acid or digestible protein and digestible amino acid basis. Feeding azolla meal on a digestible protein and digestible amino acid basis improved egg production by 5-10%, egg mass output by 3.2 -6.4 g /day and feed efficiency by 6-12 %. Feeding azolla on a digestible protein and digestible amino acid basis maintained or improved protein utilisation efficiency. The yolk colour significantly improved with increased levels of azolla meal and duration of feeding. Feed cost per kg egg mass production reduced with an increased level of azolla meal in the diet and reduced proportionally more when formulated on a digestible nutrient basis.

I. INTRODUCTION

The cost of feed may be decreased in two ways. Firstly, use of cheaper unconventional feed ingredients may help to decrease feed cost but the effect of these ingredients on poultry production needs to be tested before they can be recommended. Secondly, nutritionists have recently become interested in formulating diets on the basis of digestible protein and digestible amino acids (Rhone-Poulenc Animal Nutrition, 1989; Fernandez *et al.*, 1995) as a means of increasing the efficiency of feed utilization. It is suggested that it would be more desirable to formulate rations on a digestible nutrient basis because this would be a better indication of the relative nutritional value of the feed. It has been found that the performance of laying hens has improved when the feed formulation was based on digestible lysine and digestible methionine concentration rather than on total lysine and total methionine concentration (Bougon and Joly,1990). The present study was therefore undertaken to compare the productive and economic advantages of feeding a new ingredient, azolla meal, on a digestible protein and digestible amino acid basis compared with feeding on a total protein and total amino acid basis. Azolla is an aquatic fern found abundantly in ponds, ditches and paddy fields in tropical and subtropical regions of the world. It contains 165-292 g /kg CP and has been found to be useful as a feed ingredient for poultry (Singh *et al.*,1983; Ali and Leeson, 1995).

II. MATERIALS AND METHODS

The fresh azolla (*Azolla pinnata*) was harvested from the Bangladesh Agricultural University poultry farm pond and then sun-dried. Azolla meal was analysed for proximate composition, acid detergent fibre (ADF) and neutral detergent fibre (NDF) using standard procedures, for metabolizable energy using the method of Sibbald and Slinger (1993) and for digestible protein using the method of McNab and Shannon (1972).

¹ Department of Animal Production, University of Queensland, Gatton College, Qld 4345.

² Department of Animal Husbandry, Hajee Md. Danesh Ag College, Bashirhat, Bangladesh.

Diets were prepared with azolla meal at a level of 50 and 100g/kg (Trial 1) and 150 and 200g/kg (Trial 2) on a total protein and total amino acid (TPTA) basis or on a digestible protein and digestible amino acid (DPDA) basis by replacing sesame meal and some whole soybean from wheat based control diets to adjust the digestible protein, digestible lysine and digestible methionine concentrations (NRC,1994; Scragg, 1994; Ali and Leeson, 1995). In trial 2, synthetic L-lysine HCl and DL- methionine were added to the diets formulated on a digestible nutrient basis to adjust the digestible lysine and digestible methionine concentrations. In trial 1, sixty laying pullets 43 weeks old, and in trial 2, eighty laying pullets (Shaver Starcross 579) 28 weeks old, were used. In trial 1, each diet was applied to three replications of 4 birds, while in trial 2 each diet was applied to four replications of 4 birds. The birds were reared in cages in an open house. All mash dry feed was supplied *ad libitum* and light was maintained at 16 h per day during the experimental periods. The experiments continued for 16 and 8 weeks for trials 1 and 2 respectively. During the experiments, daily egg production, egg weight and weekly feed consumption were recorded. In trial 1, one egg from each replicate was collected for the first three days and last three days of weeks 4, 8, 12 and 16 and in trial 2, one egg from each replicate was collected on the last three days of weeks 4 and 8 to determine egg quality. The external egg quality characteristics measured were egg weight, shape index, egg breaking strength, per cent shell and shell thickness. The internal egg quality characteristics measured were Haugh unit, albumen index, albumen dry matter, yolk colour score, yolk index, and yolk dry matter. Hen-day egg production, egg mass output, feed efficiency, protein efficiency ratio (PER) and feed cost were calculated. Data were subjected to analysis of variance and significant differences between treatments were identified by the least significant difference test.

III. RESULTS

The chemical analysis and digestibility study of azolla meal indicated that the crude protein was 285g/kg, digestible protein was 220g/kg, ash was 169g/kg, ADF was 334g/kg, NDF was 446g/kg and crude fibre was 124g/kg. The metabolizable energy of azolla meal was 10.17MJ/kg. The performance of laying hens fed azolla meal (AM) at different levels either on a total protein and total amino acid (TPTA) basis or on a digestible protein and digestible amino acid (DPDA) basis is shown in Table 1.

Table 1. Hen-day egg production, (HDEP, %), egg mass output (EMO, g/d), food intake (FI, g/d), FCR, protein utilisation efficiency (PUE, %), livability (%), egg weight (g), feed cost/kg (c), feed cost/hen/unit egg mass output (A\$) of laying hens fed azolla meal on total protein and total amino acid, or digestible protein and digestible amino acid basis.

Performance	Level of azolla meal (g/kg)									
	0		50		150		100		200	
	(Control)		(TPTA)		(DPDA)		(TPTA)		(DPDA)	
Trial	1	2	1	2	1	2	1	2	1	2
HDEP	75.7	73.5 ^{ab}	72.1	70.7 ^{bc}	82.1	73.3 ^a	72.0	69.4 ^c	77.7	73.7 ^{ab}
EMO	49.5	39.6 ^c	48.3	38.2 ^c	52.7	44.5 ^a	47.3	38.9 ^c	50.9	42.0 ^b
FI	118.4	107.3 ^c	118.6	112.6 ^{ab}	119.4	108.2 ^{bc}	118.1	113.5 ^a	119.2	107.3 ^c
FCR	2.42	2.75 ^b	2.47	2.99 ^a	2.28	2.43 ^c	2.52	2.95 ^a	2.36	2.56 ^c
PUE	28.0	25.1 ^{bc}	26.4	23.2 ^c	29.5	27.5 ^a	26.3	22.8 ^c	27.8	26.1 ^{ab}
Livability	100	100	100	100	100	100	100	100	100	100
Egg weight	63.3	55.5 ^{bc}	66.9	54.5 ^c	64.4	57.5 ^a	65.7	55.0 ^c	65.5	56.9 ^{ab}
Feed cost/kg	29.4	29.5	27.9	25.9	28.2	27.5	26.8	24.7	26.6	26.7
Feed cost/ hen	12.8	11.7	12.4	11.2	11.6	9.6	12.2	10.5	11.3	9.9

Means in the rows within trial bearing similar superscripts do not differ significantly (* P <0.05).

The hen-day egg production at 50 g and 100 g AM/kg using digestible protein and digestible amino acid (DPDA) was improved by 9.92 and 5.67 per cent respectively (Trial 1), while in Trial 2, at 150 and 200 g AMDPDA/kg, egg production improved ($P < 0.05$) by 2.58 and 4.24 per cent respectively compared with similar levels of AMTPTA. Feeding AMTPTA up to 150 g/kg maintained egg production equal to the control group but egg production reduced ($P < 0.05$) at 200 g/kg diet. The egg mass output (g egg/day) at 50 and 100 g AMDPDA/kg improved by 4.43 g and 3.59 g (Trial 1), and at 150 and 200 g AMDPDA/kg improved ($P < 0.05$) egg mass output by 6.36 and 3.18 g (Trial 2) compared with similar levels on a AMTPTA basis. The egg mass output of birds fed the control diet, 150 or 200 g AMTPTA, was similar. Feed consumption was similar in all dietary treatments (Trial 1) but significantly increased ($P < 0.01$) for the 150 g AMTPTA (112.6 g) and the 200 g AMTPTA (113.6 g) diets above that of the control (107.3 g) diet (Trial 2). Feed consumption at 150 g AMDPDA (108.2 g) and 200 g AMDPDA (107.3 g) was similar to that of the control diet. Feed conversion ratios were found to be better for the 50 and 100 g AMDPDA diets than for the control, 50 and 100 g AMTPTA diets (Trial 1) but significantly improved ($P < 0.05$) for the 150 and 200 g AMDPDA diets compared with the control or similar levels of the AMTPTA diets (Trial 2). PER significantly ($P < 0.05$) improved at 50 g AMDPDA but reduced at 50 and 100 g AMTPTA diets compared with that of the control diet (Trial 1). The PER of the 150 g AMDPDA diet was significantly better than the PER of the control diet or the 150 g AMTPTA diet, while the PER of the 200 g AMDPDA diet was significantly better than the PER of the 200g AMTPTA diet but was similar to that of the control diet (Trial 2).

Livability was 100% in all dietary treatments in both the trials. The external characteristics of eggs such as shape index, egg breaking strength, percent shell, shell thickness, and internal egg quality characteristics such as Haugh unit, albumen index, albumen dry matter (%), yolk index, and yolk dry matter (%) of the eggs laid by hens in different treatments at starting, 4, 8, 12, and 16 weeks (Trial 1) and at 4 and 8 weeks (Trial 2) were similar except egg size and yolk colour. The egg size improved ($P < 0.01$) when feed was formulated on the basis of DPDA (Trial 2). The yolk colour significantly improved ($P < 0.05$) with increasing level of azolla meal and period of feeding whether fed on a total or a digestible protein and amino acid basis. The feed cost per hen per year at 50, 100, 150, and 200 g/kg azolla meal diet reduced by A\$ 0.40 versus 1.23, A\$ 0.65 versus 1.51 (Trial 1), A\$ 0.53 versus 2.07, and A\$1.20 versus 1.85 (Trial 2) compared with the costs of the control diets when fed on a TPTA versus DPDA basis respectively.

IV. DISCUSSION

Azolla meal was a moderate source of crude protein and metabolizable energy. The analysed crude protein of azolla meal was similar to the value reported by Singh *et al* (1983) but was greater than values reported by others (Tamang and Samanta, 1993; Ali and Leeson, 1995). Digestible protein was similar to the value reported by Querijero (1991) but was greater than that reported by Ali and Leeson (1995). The NDF and ADF content were less than reported previously in terms of total crude fibre. The metabolizable energy of azolla meal was similar to that of duck weed (Akhter, 1995) but greater than reported previously for azolla meal (Ali and Leeson, 1995). These differences indicate that the azolla meal used in these studies was of high quality. Feeding azolla meal on a DPDA basis improved egg production in laying hens, while egg production reduced at greater levels of azolla meal when fed on a TPTA basis. The results are partially consistent with Bastian (1987). These indicate that azolla meal can replace sesame meal on a DPDA basis up to 200 g/kg diet of laying hens but only up to 150 g/kg diet when fed on a TPTA basis. Feeding azolla meal on a TPTA basis

had no effect on egg mass output but output improved when birds were fed on a DPDA basis. The results confirm earlier observations of improved egg mass output in laying hens when fed on a digestible protein and digestible amino acids basis (Bougon and Joly, 1990; Joly, 1994). Feeding azolla meal at a lower level on a TPTA or on a DPDA basis had no effect on feed consumption but feed consumption increased at a higher level of azolla meal when fed on a TPTA basis. Feed efficiency improved even at 200 g/kg azolla meal when fed on a DPDA basis but reduced when fed on a TPTA basis. The results are similar to earlier observations (Bougon and Joly, 1990; Johnson, 1992; Joly, 1994) of higher feed efficiency when fed on a digestible nutrient basis. Protein efficiency ratios were maintained or improved when azolla meal was fed on a DPDA basis but reduced when fed on a TPTA basis compared with the controls.

No bird died in any treatment during the experiments, indicating that azolla meal had no deleterious effect on livability. Azolla meal had no influence on external or internal characteristics of eggs except egg size and yolk colour. Egg size improved when azolla meal was fed on a DPDA basis. Yolk colour improved with increased levels of azolla meal in the diets and for longer periods of feeding whether fed on a TPTA or a DPDA basis. Feed cost reduced with increasing level of azolla meal in the diet and reduced more when fed on a digestible protein and digestible amino acid basis than on a total protein and total amino acid basis.

V. CONCLUSION

Egg production, feed efficiency and profitability increased when azolla diets were formulated on a digestible protein and digestible amino acid basis compared with the usual formulation on a total protein and total amino acid basis.

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NUTRITIVE VALUE OF LUPINS FOR LAYERS

R.J. HUGHES and A. KOCHER

Summary

Nutritive values of Australian sweet lupin *Lupinus angustifolius* cv. Gungurru and white lupin *Lupinus albus* cv Kiev mutant were assessed in a series of three experiments each of eight weeks duration which were conducted on the same flock of 960 Tegel Tint and SB2 hens.

Layer diets containing 28% wholeseed Gungurru, 19% de-hulled Gungurru, 18% wholeseed Kiev or 15% de-hulled Kiev produced comparable results in terms of rate of lay, egg size, excreta condition and egg quality. The seed coat of both species was deemed to be low in energy but had no obvious anti-nutritive effects in diets for laying hens. Commercial enzyme product reduced feed intake and incidence of soiled eggs. In another experiment, dietary inclusion of 22.5% wholeseed Gungurru in wheat or sorghum-based diets had no deleterious effects on feed intake, rate of lay, excreta moisture or egg quality in comparison with control diets containing no lupin. However, an experiment with a commercial isolate of NSP from de-hulled *L. angustifolius* showed that there was potential for wet droppings and increased soiling of eggs as a result of high dietary levels of NSP from Gungurru lupin.

In conclusion, Australian sweet lupin and white lupin are valuable alternative sources of protein for inclusion in layer diets. Removal of seed coat is an unnecessary added cost but care needs to be taken when assigning energy values to lupin for formulation purposes. Further development of enzymes is required for removal of anti-nutritive effects of NSP from lupin.

I. INTRODUCTION

High protein, amino acid and energy levels, combined with cost-competitiveness with a wide range of cereals, legumes and animal proteins, give Australian lupins excellent potential for use in poultry diets. *Lupinus angustifolius* cv. Gungurru, in particular, is widely used as a protein source for monogastrics (van Barneveld and Hughes, 1994).

However, some uncertainty surrounds the effects of high levels of oligosaccharides and non-starch polysaccharides (NSP), the value of additional processing such as de-hulling, and the cost-effectiveness of commercial enzyme products. Do high levels of oligosaccharides and NSP increase the incidence of wet droppings and soiled eggs? Does the seed coat of lupins contain anti-nutritive factors and will de-hulling lead to improvements? Can addition of feed enzymes to diets containing wholeseed or de-hulled lupins improve laying performance?

Answers to these questions were sought in a series of experiments conducted on the same flock of hens. Summarised results of three experiments are discussed in this report.

II. MATERIALS AND METHODS

A total of 960 laying hens (480 Tint and 480 SB2) were housed five per cage in 192 Harrison "Welfare" back-to-back, single-tier cages (each 500 mm wide by 545 mm deep; 545 cm²/bird) in a controlled environment layer shed. A 16-hour light program was provided. Hens had access to feed and water at all times. From 18 to 36 weeks of age, and in periods between experiments, birds received a high nutrient density layer mash.

SARDI, Pig and Poultry Production Institute, University of Adelaide, Roseworthy, SA 5371.

In each of the experiments, 8 dietary treatments (12 replicates; 10 birds in two adjacent cages for each replicate) were examined over an 8-week period. Dietary treatments were re-randomised for each experiment. Major feed ingredients were analysed for protein and AME (by broiler bioassay). All diets within an experiment had similar levels of energy, protein, linoleic acid, essential amino acids, Ca, available P, Na and Cl. Hens were weighed at the start and end of each experiment. Hen-day egg production and mortality were recorded daily. Feed intake was measured at 4-week intervals. Egg weight, excreta moisture, incidence of soiled eggs, shell thickness and yolk colour were measured at the end of experiments.

Experiment 1 examined the effects of lupin species (*L. angustifolius* cv Gungurru and *L. albus* cv Kiev mutant), removal of seed-coat, and dietary addition of a proprietary enzyme product (Avizyme 2300 at 1kg/tonne) in wheat-based diets on the performance of laying hens in the period 36 - 44 weeks of age. Inclusion rates of lupin were Gungurru wholeseed 27.9%, Gungurru kernel 18.9%, Kiev wholeseed 18.2%, and Kiev kernel 15.0%. Steam-pelleted diets contained ME 11.3 MJ/kg, protein 17%, lysine 0.77%, methionine 0.36%, Ca 3.75% and available P 0.38%.

Experiment 2 examined the effects of dietary inclusion rate (0, 5, 10 and 15%) of a commercial isolate of NSP from de-hulled *L. angustifolius* in a sorghum-based diet, and addition of enzyme product (Avizyme 2300 at 1kg/tonne) on the performance of laying hens in the period 59 - 67 weeks of age. The total NSP content of the isolate was 56% which was mainly galactose (75%). The soluble NSP content of the isolate was 6%. Steam-pelleted diets contained ME 11.3 MJ/kg, protein 15.5%, lysine 0.70%, methionine 0.35%, Ca 4.0% and available P 0.35%.

Experiment 3 examined the effects of dietary inclusion rate (0, 7.5, 15 and 22.5%) of *L. angustifolius* cv Gungurru in wheat or sorghum-based diets on the performance of laying hens in the period 81 - 89 weeks of age. Mash diets contained ME 11.5 MJ/kg, protein 17%, lysine 0.77%, methionine 0.36%, Ca 3.8% and available P 0.4%.

III. RESULTS AND DISCUSSION

Results from experiment 1 (Table 1) show that egg production, excreta moisture, and shell quality were unaffected by lupin species, form of lupin or enzyme addition. Gungurru lupin increased feed intake by 4.3% compared with Kiev lupin, whereas de-hulling of both species reduced feed intake by 1.7%. Enzyme addition reduced feed intake by 3.9%, egg size by 1.2% and incidence of soiled eggs by 28%. Enzyme degradation of NSP in ingredients other than lupin could have contributed to these benefits. Yolk colour was significantly reduced by de-hulling (3.3%). On the other hand, yolk colour was improved by 3.3% by addition of enzyme to the Gungurru diets. Tegel Tint hens ate more feed (4.2%) and laid more eggs (5.2%) although egg size was smaller (1.2%) in comparison with Tegel SB2 hens.

These results suggest that energy values for Kiev lupin were underestimated relative to Gungurru, as were the energy values for lupin kernel relative to whole seed of both species, possibly because AME values obtained in chick bioassays are not directly applicable for formulation of layer diets. Seed coat of both lupin species appear not to contain anti-nutritive factors but did have an energy dilution effect in this study.

In experiment 2, dietary inclusion of lupin kernel isolate reduced feed intake and yolk colour, and increased excreta moisture content. However, there was no evidence of a dose response effect of lupin NSP on feed intake or excreta moisture. On the other hand, yolk colour was significantly affected by an interaction between breed and lupin NSP, with higher levels of lupin NSP depressing yolk colour in Tint but not in SB2 hens. Addition of enzyme had no significant effects on performance, excreta condition or egg quality.

Table 1. Effects of breed (SB2 and Tint), lupin species (Gungurru and Kiev), lupin form (wholeseed and de-hulled) and enzyme product on feed intake (FI), egg production (EP), excreta moisture (EM), soiled eggs (SE), egg weight (EW), shell thickness (ST) and yolk colour (YC). Non-significant effects ($P>0.05$) are shown as ns.

Effect	FI (g/bird/d)	EP (%)	EM (%)	SE (%)	EW (g)	ST (μ m)	YC (Roche)
<i>Probability of greater F value in analysis of variance</i>							
Breed	<0.001	ns	ns	ns	0.06	ns	ns
Lupin	<0.001	ns	ns	ns	ns	0.08	0.09
Form	0.06	ns	ns	ns	ns		<0.001
Enzyme	<0.001	ns	ns	0.06	<0.05	ns	ns
<i>Least squares means</i>							
SB2	110.0	82.0	76.8	8.7	60.4	372	9.0
Tint	116.6	86.5	76.3	7.8	59.7	371	9.1
Gungurru	116.5	83.9	76.5	9.2	60.0	374	9.1
Kiev	111.1	84.6	76.6	7.3	60.1	369	9.0
De-hulled	112.9	84.2	76.8	8.9	59.9	372	8.9
Wholeseed	114.8	84.3	76.2	7.6	60.3	371	9.2
No enzyme	116.0	83.8	77.0	9.6	60.4	372	9.0
Enzyme	111.6	84.7	76.1	6.9	59.7	371	9.1
Mean	113.8	84.3	76.6	8.6	60.0	371	9.0

Table 2. Effects of breed (SB2 and Tint), dietary inclusion rate (0, 5, 10 and 15%) of a commercial isolate of NSP from *L. angustifolius*, and enzyme product on feed intake (FI), egg production (EP), excreta moisture (EM), soiled eggs (SE), egg weight (EW), shell thickness (ST) and yolk colour (YC). Non-significant effects ($P>0.05$) are shown as ns. Lupin NSP mean values having a common superscript are not significantly different ($P<0.05$).

Effect	FI (g/bird/d)	EP (%)	EM (%)	SE (%)	EW (g)	ST (μ m)	YC (Roche)
<i>Probability of greater F value in analysis of variance</i>							
Breed	<0.001	<0.05	<0.001	<0.001	<0.05	<0.05	<0.01
Lupin NSP	<0.05	ns	<0.001	ns	ns	ns	<0.001
Enzyme	ns	ns	0.06	ns	ns	ns	0.07
<i>Least squares means</i>							
SB2	120.3	69.5	78.7	16.0	66.2	366	9.0
Tint	114.9	73.1	80.5	8.9	65.1	359	8.7
Lupin NSP 0%	121.8 ^a	72.4	77.4 ^a	12.4	65.8	366	8.9 ^b
Lupin NSP 5%	116.7 ^b	72.3	79.8 ^b	11.2	65.0	357	9.2 ^a
Lupin NSP 10%	114.9 ^b	68.1	80.1 ^b	13.3	65.9	364	8.7 ^{ab}
Lupin NSP 15%	117.2 ^b	72.4	81.0 ^b	12.9	65.8	364	8.6 ^c
No enzyme	117.4	71.9	79.1	13.2	65.5	364	8.8
Enzyme	117.9	70.7	80.1	11.7	65.8	362	8.9
Mean	117.08	70.8	75.9	12.6	65.6	363	8.9

In experiment 3, inclusion of up to 22.5% Gungurru lupin in wheat or sorghum-based diets had no deleterious effects on laying performance, excreta condition, or egg quality (Table 3). Yolk colour was improved by at least 0.2 Roche fan units at higher levels of inclusion. Significant increases in feed intake (2.8%) and incidence of soiled eggs (29%) by hens given sorghum-based diets was unexpected. However, this might be a characteristic of this particular sample of sorghum which had lower than usual AME and promoted wetter than usual droppings in broiler chickens in AME bioassays (Hughes, unpublished data).

Table 3. Effects of breed (SB2 and Tint) and dietary level (0, 5, 10 and 15%) of NSP isolated from *L. angustifolius* cv Gungurru in wheat or sorghum-based diets on feed intake (FI), egg production (EP), excreta moisture (EM), soiled eggs (SE), egg weight (EW), shell thickness (ST) and yolk colour (YC). Non-significant effects ($P>0.05$) are shown as ns. Lupin mean values having a common superscript are not significantly different ($P<0.05$).

Effect	FI (g/bird/d)	EP (%)	EM (%)	SE (%)	EW (g)	ST (μ m)	YC (Roche)
<i>Probability of greater F value in analysis of variance</i>							
Breed	<0.001	ns	0.07	ns	ns	<0.05	ns
Lupin level	ns	ns	ns	ns	ns	0.06	<0.001
Grain base	<0.05	ns	ns	<0.05	ns	ns	ns
<i>Least squares means</i>							
SB2	122.2	51.4	70.1	10.0	65.4	345	11.8
Tint	107.3	52.7	71.2	10.3	65.6	336	11.7
Lupin 0%	112.9	51.8	70.6	6.9	64.9	348	11.6 ^c
Lupin 7.5%	115.7	52.4	70.7	10.6	66.7	336	11.6 ^{bc}
Lupin 15%	115.2	51.4	70.5	11.7	65.8	344	12.0 ^a
Lupin 22.5%	115.3	52.6	70.9	11.5	64.7	335	11.8 ^{ab}
Sorghum	116.4	52.2	70.7	11.9	66.0	341	11.7
Wheat	113.1	51.8	70.6	8.4	65.1	340	11.8
Mean	114.8	52.0	70.6	10.1	65.5	342	11.8

IV. CONCLUSIONS

Australian sweet lupin and white lupin are valuable alternative sources of protein for inclusion in layer diets. The seed coat of neither species contained anti-nutritive factors but did have an energy dilution effect. It is possible that beneficial effects from the enzyme product used in these studies came from degradation of NSP from other ingredients as well as lupin.

V. ACKNOWLEDGMENTS

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USING LABORATORY STUDIES TO IMPROVE THE IN FIELD APPLICATION OF *EIMERIA SPECIES*

D.G. RICHARDS

Summary

Four groups of commercial pullets were dosed at 3 days of age with a field strain of *Eimeria necatrix*. Oocysts were administered to each bird within each group at 15, 75, 150 and 1500 oocysts *per oris* respectively. The group dosed with 1500 oocysts produced an oocyst output with a reproductive index of 28 while the group dosed with 15 oocysts produced an oocyst output with a reproductive index of 9515. The groups dosed with 75 and 150 oocysts per bird had intermediate reproductive indexes. Birds receiving 1500 oocysts at 3 days had significantly ($p < 0.01$) reduced growth rates between 3 and 14 days when compared to each control. All groups dosed with oocysts at 3 days of age and challenged with 1500 oocysts of the same strain at 18 days of age showed lowered reproductive index. Between 3 and 14 days of age there was a significant ($p < 0.01$) difference in body weight gain between the undosed / unchallenged bird group and the group dosed with 1500 oocysts. For the 14-26 age period, the undosed / challenged group, the 75 oocyst and 1500 oocyst group had significantly ($p < 0.01$) lowered body weight gain when compared to the undosed / unchallenged control. Between 14 and 26 days of age there were no significant ($p < 0.01$) differences in body weight gain between the challenged bird group and the dosed / challenged groups. The results demonstrate the effects of crowding with pathogenic strains of coccidia, examine age susceptibility and establish a model to be used to compare the immunogenicity and pathogenicity of Australian strains of *Eimeria*.

I. INTRODUCTION

Seven strains of *Eimeria* have been isolated in Australia, with *Eimeria mitis* and *E. praecox* being the most recent (Jorgensen *et al*, 1997). In the Australian poultry industry, *E. acervulina*, *E. maxima*, *E. tenella* and *E. acervulina* are regarded as the most common and pathogenic. The 2 strains the author has observed as most pathogenic in flocks of broiler breeders are *E. tenella* and *E. necatrix*. Attenuated strains of *E. acervulina* and *E. maxima* have been developed in Australia (Jorgensen and Stewart, 1996) and these are undergoing assessment for incorporation into a live vaccine. Work is continuing on attenuating *E. tenella* and *E. necatrix*.

To stimulate immunity in commercial flocks the poultry industry uses a program of controlled exposure. This approach requires administration of live *Eimeria* oocysts, usually to birds less than 10 days of age, followed by strategic application of water-soluble anticoccidials. However there is little information on the efficacy of the dose rates used. The following experiment summarizes the dose response trial undertaken with a field strain of *E. necatrix* used in controlled exposure programs. The aim is to use the trial format to develop a model to enable species comparisons.

Medichick Pty Ltd, 29 Tantani Street, Frankston North, Victoria, 3200.

II. MATERIALS AND METHODS

(a) Birds, feed and body weights

Day old ISA pullets were individually identified and randomly placed into 5 groups. Birds were fed *ad libitum* on a commercial feed which was coccidiostat free (Barastoc Ostrich Starter, Ridley Agriproducts). Water was not restricted. Pen litter was replaced daily. Trial birds were brooded as 5 groups of 11 birds (4 treatment groups and 1 control) as well as a pooled group of birds from the same hatch. The pooled birds were used for in-contact transmission determination. In-contact birds were placed with each dosed group of birds between 10 and 16 days post vaccination. At 16 days, in-contact birds were postmortemed to determine the degree of oocyst residual infection in the brooding boxes. The estimation used gut lesion scoring. At 15 days post vaccination, the control group was split into 2 rearing boxes. One group was challenged with 1500 oocysts (undosed / challenged). The 4 groups dosed at 3 days were challenged with 1500 oocysts at the same time. The remaining group of birds was described as undosed \ unchallenged and "challenged" with 1 mL of phosphate buffered saline (PBS). Birds were weighed on an electronic balance on days 0,3,8,11,14,18,23 and 26. In-contact birds were weighed on placement with infected birds and prior to lesion score assessment.

(b) Eimeria necatrix culture

Oocysts obtained from a laboratory strain (MCK strain) were counted. A 1-mL sample of oocyst suspension (stored in PBS to which 0.1% formalin had been added) was mixed with 49 mLs of saturated sodium chloride and gently agitated. A 0.5-mL aliquot collected from mid-tube was placed under a McMaster counting chamber and left for 5 minutes. Four samples were counted and the mean calculated. The culture concentration was corrected for sporulation rate by multiplying the count by the percentage sporulation rate. Serial dilutions for individual bird administration were carried out using PBS as the diluent. At 3 days of age, each of the 4 treated groups of birds were inoculated into the oesophagus using a syringe with 1500, 150, 75 and 15 oocysts contained in 1 mL of PBS. The control group birds were given 1 mL of PBS in the same way. The "pooled" in-contact birds were untreated. At 18 days of age, the 1500, 150, 75, 15 dose groups and one untreated group of birds were challenged with 1500 oocysts of the MCK strain from the same culture used at 3 days of age.

(c) Faecal sampling and counting

From 6 to 9 days post dosing and 6 to 9 days post challenge, grids were placed into the rearing boxes to catch faeces. The faeces were weighed and mixed in a large plastic bag. A 300 GM sample or the total weight (if the faecal sample weighed under 300 GM) was made up to a volume of 1000-mL using water. The sample was then homogenised for 30 seconds using a hand held kitchen chopper. From each sample 4 individual 5-mL samples were collected using a coarse tip pipette and placed into a tube containing 45 mL of saturated sodium chloride. The tubes were capped and gently agitated, after which 4 by 0.5-mL samples were placed under a McMaster counting chamber, left for 5 minutes and the oocysts counted.

III. RESULTS

Results of the oocyst output per group were expressed as a reproductive index (RI): which is the total oocyst output divided by total oocyst dose. The RI results are on table 1. The contact bird lesion scores are located at the foot of table 1. The body weight gains are summarised on Table 2.

Table 1 Reproductive index (RI) and lesion scores in birds dosed with *Eimeria necatrix* (MCK strain) at 3 days of age and challenged at 18 days of age.

Group	Treatment at 3 d/o	RI. 6-9 days	Treatment at 18 d/o	RI. 23-26 days
1	15 oocysts	9515	1500 oocysts	4.11
2	75 oocysts	2163	1500 oocysts	0.96
3	150 oocysts	3260	1500 oocysts	25.00
4	1500 oocysts	28	1500 oocysts	4.71
5	0 oocysts	0	1500 oocysts	267.00
6	0 oocysts	0	0 oocysts	0.00

Post mortem of the four in-contact birds placed with groups 1,2,3,4 and 5 for days 11 to 16: No lesions of *E necatrix* were observed in any in-contact bird.

Table 2 Mean body weight gains BWG (GM / bird) for 3-14 and 14-26 days of age.

Group	Treatment at 3 d/o	BWG 3-14 d	Treatment at 18 d/o	BWG 14-26 d/o**
1	15 oocysts	98 ^a	1500 oocysts	157 ^a
2	75 oocysts	86 ^a	1500 oocysts	124 ^b
3	150 oocysts	98 ^a	1500 oocysts	155 ^a
4	1500 oocysts	72 ^b	1500 oocysts	148 ^c
5	0 oocysts	94 ^a	1500 oocysts	128 ^d
6	0 oocysts	94 ^a	0 oocysts	166 ^a

BWG with differing superscripts for 3-14 d are significantly different to group 6 at $p < 0.01$

BWG with differing superscripts for 14-26 d are significantly different to group 6 at $p < 0.01$

**There is no significant difference ($p < 0.01$) between groups 1,2,3,4 and 5.

IV. DISCUSSION

Birds dosed with oocysts at 3 days of age and challenged 15 days later had reduced oocyst output on the second cycle. Birds receiving a primary dose of 1500 oocysts at 3 days had lowered oocyst output when compared to the group receiving the same dose at 18 days of age. The reduction in reproductive index with increasing age is consistent with the "crowding" effect noted by other authors (Fernando, 1982). Each group dosed with oocysts at 3 days of age had the ability to suppress oocyst output when challenged with 1500 oocysts 15 days later. This shows older birds receiving the same dose had increased oocyst output. Similar trends have been observed where oocyst production was higher in older birds (Fernando, 1982). However, there is a report where a single dose of 80,000 oocysts of *E*

necatrix resulted in no survivors. In the author's laboratory, 4,000 oocysts of the strain of *E necatrix* used in this trial caused mortality in birds under 2 weeks of age. This could indicate the strains of *E necatrix* in Australia are more pathogenic than those reported in overseas literature.

The group dosed with 1500 oocysts at 3 days had significantly reduced growth rate throughout the trial. Birds dosed for the first time with 1500 oocysts at 18 days of age had significantly reduced growth rates between 14 and 26 days. The group dosed with 75 oocysts and challenged with 1500 oocysts also had a significantly reduced growth rate between 14 and 26 days. The dose of 15 and 150 oocysts produced no significant body weight gain depression during the trial period. Both of these doses produced lowered oocyst output. There are no known reasons why the 75-oocyst group produced a lower body weight gain difference to the unchallenged controls between 14 and 26 days. It is to be noted this group produced a reduction in oocyst output during the second cycle, which indicates an immune response was operating. The body weight depression with the 75-oocyst dose does not fit the trend established by the 15 and 150-oocyst dose, and requires repeating.

Under field conditions, oocysts recycle. This recycling builds up oocyst numbers in the shed. It is the ingestion of oocysts in uncontrolled numbers which causes mortality. This trial shows 15 oocysts of a field strain of *Eimeria necatrix* can stimulate a bird's immune system and afford some protection to 1500 oocysts within 15 days. The effect a 15-oocyst dose has on the bird's ability to withstand challenge, as the result of oocyst re-exposure, is not demonstrated in this trial. In controlled exposure programs, it is important to ensure all birds receive a minimum immunising dose. This may be particularly important where species used are pathogenic. One consideration with pathogenic species is their application. Fernando (1982) reported studies in birds dosed with *E necatrix* at a single dose of 20,000 oocysts, compared with the same dose split over 4 lots of 5,000 oocysts. The single dose was more pathogenic and caused mortality. As the Australian strains of *E necatrix* used in controlled exposure programs are pathogenic, the application frequency used needs investigating. An application spread over 4 days will reduce the rate of second cycle oocyst build up which could reduce the chances of observing pathogenic effects.

With reference to the controlled exposure doses administered to birds by the poultry industry, the dose of *Eimeria necatrix* used is as low as 25 oocysts. Further trials are required to re-examine the 15 to 75 oocyst dose range and study the effects of oocyst recycling under single and repeated administration patterns. This trial indicates a dose of 25 oocysts should be suitable as a primary dose. If species of *Eimeria* are to be compared, and in order to remove the effects of age, dose response trials should follow the format and bird ages used in this study.

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MORTALITY PATTERNS IN AUSTRALIAN AND IMPORTED LAYING HENS

R.B. CUMMING, W. BALL and J.V. NOLAN

Summary

The mortality patterns in two Australian and four imported strains of egg layers, involving a total of over 3,500 birds, are presented. Commercial one-day-old pullets were obtained from hatcheries where they received routine vaccinations. Subsequently the birds were reared on a commercial farm near Tamworth, where they received typical commercial management. At 16–18 weeks of age, they were transferred to the University of New England and housed in five- or two-bird cages.

Mortalities from Marek's Disease and cannibalism complex were substantially higher in the imported strains. Mortality in the imported strains from cannibalism complex was significantly higher in five- than two-bird cages and the ranking of strains was influenced by housing system.

I. INTRODUCTION

The recent importation of laying strains from the northern hemisphere into Australia has led to a number of problems concerning bird livability. The imported strains, when vaccinated with conventional Australian Marek's vaccines, do not appear to be well protected against Marek's Disease (MD) and heavy losses have been reported in the field. A number of reasons have been put forward, including the presence of very velogenic strains of MD.

This report details a total mortality survey of two Australian and four imported strains of laying birds to 66 weeks of age under management conditions that are typical of many Australian poultry farms. The paper by Nolan et al. (1998) in these proceedings details the production and economic traits of this trial.

II. MATERIALS AND METHODS

The birds were hatched between 16 and 30 January 1996. All chickens were vaccinated at their hatcheries against MD and Infectious Bronchitis (IB) at one-day-old. They were reared in wire floored cages on a large commercial farm near Tamworth where they received a conventional chicken starter crumble diet to eight weeks of age, a crumbled grower diet from 8 to 18 weeks of age, and a crumbled pre-layer diet from 18 to 22 weeks of age. At three weeks of age, the birds were re-vaccinated against IB (A3 virus – in contact method) and at 14 weeks against Avian Encephalomyelitis and IB (Vic S – in contact method). The birds were beak trimmed at ten days of age and again at eight weeks of age.

The birds were moved when 17–18 weeks of age to Laureldale Poultry Farm, University of New England, where they were housed in single deck laying cages at either five birds per cage (modern cages, Shed 1) or two birds per cage (Californian cages, Shed 2). Shed 1 contained 440 birds in 88 cages (five birds/cage) of each of the Hy-Line CB, Isa Brown, Tegel Black and Hisex strains, while the Lohmann Brown and Hy-line Brown strains were represented by 480 birds each. Shed 2 had 132 birds per strain in two-bird cages. Birds that died were replaced up to 22 weeks of age.

Department of Animal Science, University of New England, Armidale, NSW. 2351.

A macroscopic post mortem examination was made on all birds that died up to 66 weeks of age. Deaths were categorised according to the most obvious abnormality or lesion when examined. Features diagnostic for MD included growths involving the ovaries, spleen, kidneys, liver and heart muscle, as well as the typical thickening of the brachial and sciatic nerves. Hens dying from MD were almost invariably out of production.

Cumming (1974) suggested that cannibalism was closely associated with salpingitis/peritonitis, and Jordan and Pattison (1996) recently made the same suggestion. The conditions included in cannibalism/cannibalism complex are prolapse, vent peck, cannibalism, and salpingitis/peritonitis. These conditions can be described as follows:

- a) *prolapse*, where the cloaca was everted, sometimes containing an unlaidd hard-shelled egg, the tissues engorged with blood and showing signs of pecking.
- b) *vent-peck*, where the cloaca was usually damaged and contused with portions of the reproductive and/or digestive tract sometimes missing. Such birds are often anaemic.
- c) *cannibalism*, where a portion of the body, usually the back and thighs had been eaten away.
- d) *salpingitis* and peritonitis, where there were macroscopic signs of inflammation of the oviduct and/or the peritoneal cavity. This condition varies from acute to chronic and the lesions vary accordingly. In the acute cases, the ovary is active and the prominent lesion is marked venous congestion of the ovary and oviduct, which usually contains small (1–4 ml) flockules of white to yellowish pus. There may be similar flockules of pus in the peritoneal cavity as well. In the chronic form, the bird is generally emaciated, the ovary atrophied and the oviduct distended with concentric layers of inspissated pus. Hens may die showing symptoms varying from the acute to the chronic form.
- e) *nephritis* – due to damage in the vent area occluding the terminal portion of the ureters.

Usually the birds in categories a, b and c were in full production, as indicated by their comb development and ovarian activity.

III. RESULTS

Cumulative mortality from 18 to 66 weeks of age in the six strains housed in the two- or five-bird cages are shown in the three figures. MD mortality is shown in Figure 1, cannibalism complex mortality in Figure 2, and total mortality in Figure 3. As can be seen from the figures, the two categories of MD and cannibalism/cannibalism complex accounted for about 90% of the total mortalities.

As shown in Figure 1, losses from MD generally began early, and peaked around the 25–35 week period. MD losses were substantially higher in the imported than local strains with essentially similar mortality and ranking of the strains between the two housing systems.

The losses from cannibalism complex (Figure 2) tended to occur later, significant losses starting at about 25 weeks of age. Again, mortality from this cause was considerably higher in the imported than local strains. There was a difference between the two housing systems in the level and pattern of mortality from this cause, and in the relative ranking of the strains. Overall, cannibalism mortality was higher in the five-bird cages and continued to increase over the laying period compared to the two-bird cages, where mortality tended to plateau in all strains at about 40 weeks of age.

Within the imported strains, there was a marked change in ranking in the two housing systems, with the Lohmann birds showing the highest cannibalism-related mortality in the five-bird cages, but low to moderate absolute and relative mortality in the two-bird cages.

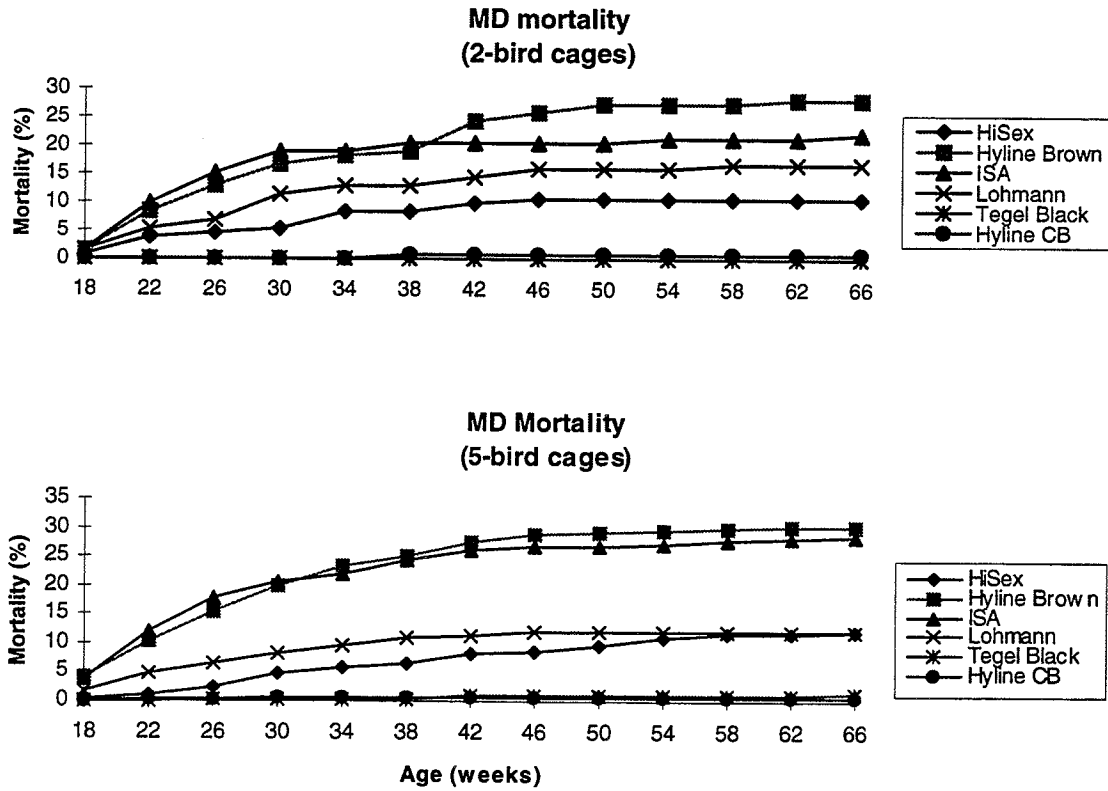


Figure 1 Cumulative Marek's Disease mortality (%) from 18 to 66 weeks of age in the six strains of layers housed in 2- or 5-bird cages.

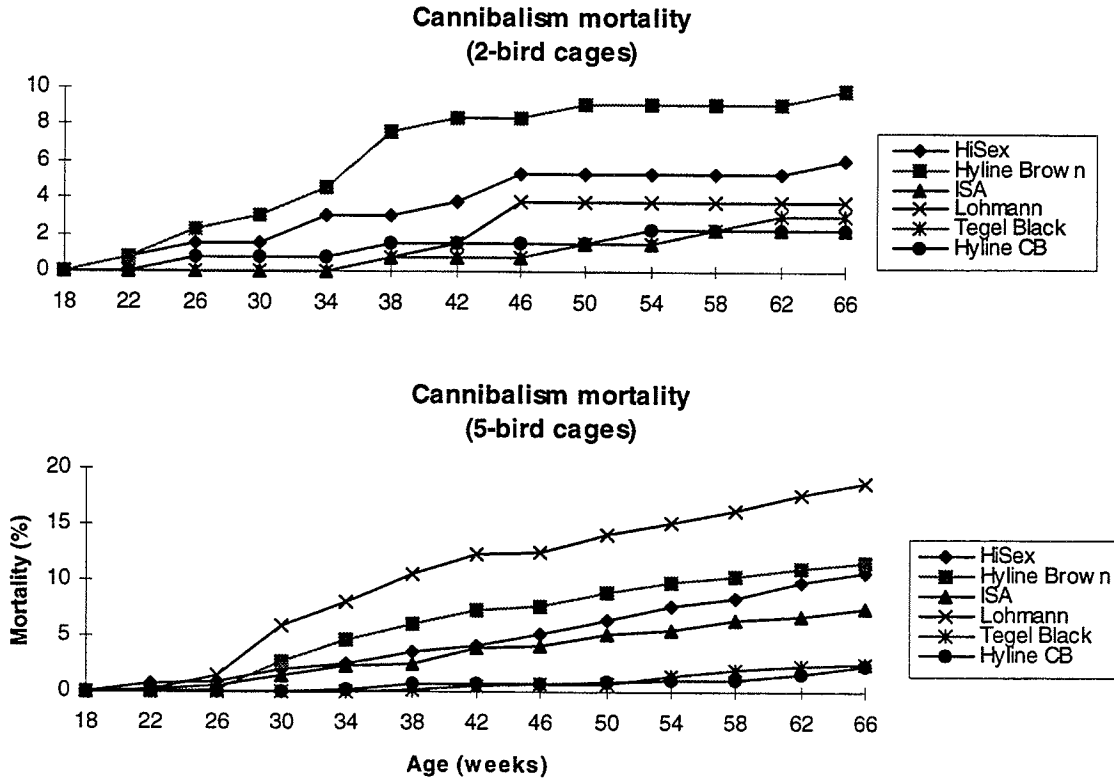


Figure 2 Cumulative cannibalism complex mortality (%) from 18 to 66 weeks of age in the six strains of layers housed in 2- or 5-bird cages.

Overall, the total mortalities in the imported breeds were considerably higher than those recorded in the Australian strains, with somewhat higher mortalities occurring in the five- than two- bird cages, in some strains at least.(Figure 3)

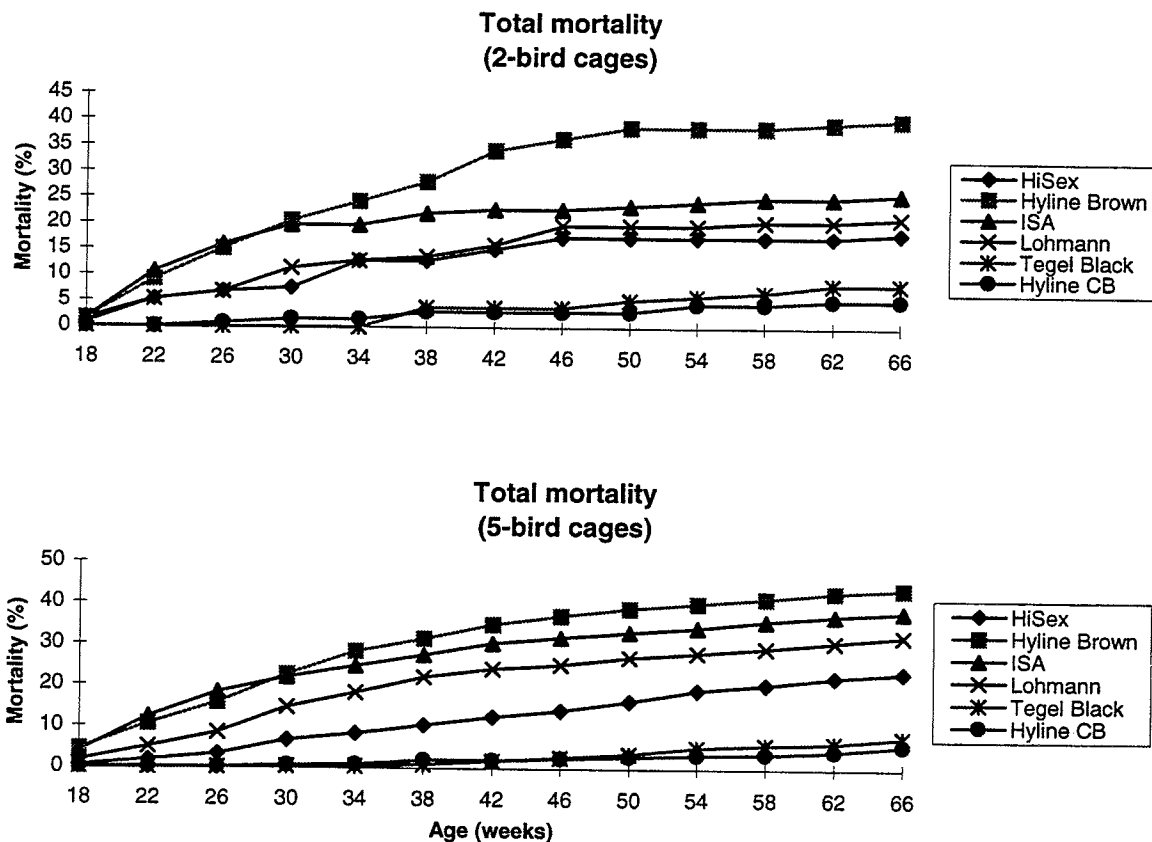


Figure 3 Cumulative total mortality (%) from 18 to 66 weeks of age in the six strains of layers housed in 2- or 5-bird cages

IV. DISCUSSION

There was no evidence of very velogenic MD strains in this trial as the two Australian strains recorded less than 2% mortality from this condition.

The cannibalism mortality results suggest that husbandry procedures such as beak trimming and lighting in sheds, which appear to be adequate for Australian strains, may need to be modified to accommodate the generally increased susceptibility of the imported strains.

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AFLATOXIN DECREASES IMMUNITY AND STRESS REACTIONS IN POULTRY

P. DEO¹, B. BLANEY² and J. DINGLE¹Summary

Pure aflatoxin B1 (AFB1) was added at a level of 0.9 ppm to the feed of chickens to monitor its effect on production, immunity and behaviour. Chickens that were fed with AFB1-contaminated diets showed a significant decrease in body weight and feed consumption. Delayed skin hypersensitivity, heterophil/lymphocyte ratio, and the size of the Bursa of Fabricius and thymus glands were also significantly decreased. Tonic immobility, a behavioural reaction to stress, was also significantly less in birds fed with AFB1.

I. INTRODUCTION

Aflatoxin (AF), particularly AFB1, has been reported to suppress growth rate and feed conversion efficiency and cause profound changes in chickens, including fatty livers and other physiological effects (Smith and Hamilton, 1970). It is the most potent hepatocarcinogen known and is capable of inducing liver cancer in most animal species (Hsieh, 1985). In addition, AF has been implicated in the induction of neoplasms in the glandular stomach, kidney, lung, salivary gland, lachrymal gland, colon and skin of domestic animals (Sharma and Salunkhe, 1991).

AFB1-induced immunosuppression has been demonstrated in domestic animals, chickens and turkeys as well as in laboratory animals (Pestka and Bondy, 1990; Sharma, 1993). Pier and McLoughlin (1985) summarised the effects of AFs on the animal immune system as: AFs impair immunogenesis without suppressing antibody formation; they suppress formation of non-specific humoral substances related to resistance and immunity (complement and interferon) and suppress phagocytosis by macrophages; they cause thymic aplasia and suppress cell mediated immunity (CMI), notably "delayed cutaneous hypersensitivity"; lymphoblastogenesis and leucocyte migration are also variably suppressed.

There are interactions between the immune system, the nervous system and the endocrine system of the body, although as yet, these relationships are not well understood (Greenberg *et al.*, 1992). Consequently, there is a great deal of interest in the parameters that control these interactions, the mediators involved and the extent to which health is modified by behaviour and vice versa. In domestic poultry, Faure (1982) uncovered a relationship between open-field activity and experimental coccidial infection in chickens selected on the basis of emotional reaction tests for fearfulness. The more fearful line showed more pronounced reactions to infection than their less fearful counterparts. Likewise, Jones (1989) found significant intra-individual correlations between the duration of tonic immobility fear reactions and leucocytic responses (heterophil/lymphocyte ratios) to stress, suggesting that leucocytic responses to chronic stressors may be greater in fearful pullets than in less fearful birds.

In general it has been found that the rank order of birds in terms of their reactions to stress and fear stays constant irrespective of the treatment. It has not yet been shown that a change in one reaction can affect a change in another. In this trial the effects of aflatoxin on poultry immune reactions and on the tonic immobility reaction to stress are examined.

¹ Department of Animal Production, The University of Queensland, Gatton, Qld 4345.

² Queensland Dept. Primary Industries, Animal Research Institute, Yeerongpilly, Qld 4105.

II. MATERIALS AND METHODS

Two basal diets were used; one formulated using sorghum, barley and soybean meal (UFFF, 1986), and one in which 150 g peanut meal/kg was substituted for some of the soybean meal. These diets were fed with or without 0.9 ppm isolated AFB1 (ICN, Australia). The AFB1 was dissolved in chloroform and then made up to 2 L in a volumetric flask. The solution was divided into two equal fractions and each half was mixed with 1.5 kg of one of the dried basal diets using a spray can. The mixture was left overnight in a forced draught fume cupboard before mixing with the remaining diet. The treatments that had 0 ppm AFB1 were mixed with chloroform only.

One week old broiler chickens were randomly allocated to single cages (8 chickens per treatment) in a temperature-controlled room ($30^{\circ} \pm 1^{\circ}\text{C}$). The birds were fed *ad libitum*. They were weighed at 7, 14 and 21 days of age. Body weight (BW), feed intake (FI) and feed conversion ratio (FCR) were calculated.

After venipuncture, one drop of blood from birds in half the replicates in each treatment was placed on a microscope slide. The blood was smeared using another slide and stained using Wright's stain. One hundred leucocytes, including heterophils, lymphocytes, monocytes, basophils and eosinophils were counted on the slide and the heterophyl/lymphocyte (H/L) ratio was calculated by dividing the number of heterophils by that of lymphocytes.

After a three day recovery from blood sampling, delayed type hypersensitivity (DTH) was measured using a procedure adapted from Kadian *et al.* (1988). Birds from half the replicates in each treatment were sensitised with 0.25 ml of 2,4-dinitrofluorobenzene (DNFB) (10mgml^{-1}) in a carrier containing acetone and olive oil (4:1) mixture. The mixture was applied to a marked area of skin on the left side of the abdomen. The right side was left as the control on which the same amount of carrier only was applied. After one week of sensitisation, the birds were challenged by DNFB (1mg ml^{-1}). The skin thickness after 24 hours was measured by a dial type micrometer (Schanelltaster, W.Germany).

Tonic immobility (TI) was measured by the method of Jones (1989) with a slight modification. Each bird was moved to a quiet room and tested individually. The bird was placed on its back on soft cloths in a box, covered and restrained by hand for 60 seconds. The observer sat nearby within sight of the bird and recorded the following; the number of inductions (Stage 1), first movement in the bird, and the duration of TI until the bird rights itself (Stage 2).

At the end of the trial the thymus lobes and the Bursa of Fabricus were removed and weighed. Their weights relative to the body weights of the chickens were analysed.

The differences between the means for BW, FI, FCR, H/L, DTH, TI and relative bursa and thymus weights for each treatment were tested for significance by the analysis of variance procedure.

III. RESULTS AND DISCUSSION

The body weight (BW), two week feed intake (FI) and feed conversion ratio (FCR) for the two week trial are tabulated in Table 1. Birds that were fed diets with 0.9 ppm pure AFB1 had significantly lower ($P < 0.001$) BW and FI. There was no significant difference in the FCR between the treatments. This results are in agreement with Reddy and coworkers (1982) who found that feeding up to 0.75 ppm and higher amounts of AF in the diet significantly depressed the weight gain and feed consumption of poultry.

Table 1. Effects on total body weight (BW), feed intake (FI), feed conversion ratio (FCR) and tonic immobility (TI) of birds fed aflatoxin supplemented diets.

Treatment	BW (g)	FI (g)	FCR	Tonic immobility (TI)	
				Day 5 (s) ¹	Day 10 (s)
Control Diet	623.7 ^{a2}	748.6 ^a	1.44	132.7 ^a	136.3 ^a
Control Diet+0.9 ppm AFB1	499.6 ^b	597.7 ^b	1.46	88.7 ^b	86.3 ^b
Peanut Diet	616.7 ^a	754.3 ^a	1.52	125.7 ^a	126.7 ^a
Peanut Diet+0.9 ppm AFB1	476.6 ^b	552.2 ^b	1.58	86.2 ^b	80.3 ^b
Significance	P<0.001	P<0.001	NSD ³	P<0.001	P<0.001
LSD	51.1	52.7	-	12.0	11.3

¹s = seconds; ²Means in a column accompanied by different superscripts differ significantly (P<0.05); ³NSD = No significant difference.

Table 2. Effects on delayed type hypersensitivity (DTH), heterophil/lymphocyte (H/L) ratio, and bursa and thymus weight of birds fed aflatoxin supplemented diets.

Treatment	DTH (mm)	H/L ratio	Bursa weight (g/100g BW)	Thymus weight (g/100g BW)
Control Diet	1.3 ^a	1.03 ^a	2.49 ^a	0.32 ^b
Control Diet + 0.9 ppm AFB1	0.6 ^b	0.81 ^b	2.26 ^{ab}	0.14 ^c
Peanut Diet	1.4 ^a	1.03 ^a	2.48 ^a	0.39 ^a
Peanut Diet + 0.9 ppm AFB1	0.7 ^b	0.82 ^b	1.95 ^b	0.13 ^c
Significance	P<0.001	P<0.01	P<0.03	P<0.01
LSD	0.2	0.12	0.33	0.08

Means in a column accompanied by different superscripts differ significantly (P<0.05).

Delayed type hypersensitivity and heterophil/lymphocyte ratios were also significantly (P< 0.01) decreased in birds that were fed AFB1 containing diets. The percentage of heterophils to total leucocytes in the blood decreased in the birds that were fed on AFB1 diets. Chang and Hamilton (1979) found that chickens with aflatoxicosis exhibited a reduction in the percentage of heterophils carrying out phagocytosis. The average weight of each thymic lobule and of the Bursa of Fabricus were also significantly (P< 0.05) less in birds fed the AFB1 containing diets (Table 2). The reduction in relative weight was greater for the thymic lobules than for the bursae. The reduction in the relative size of the Bursa of Fabricus and thymic lobules was initially observed by Thaxton *et al.* (1974) and later by Kubena *et al.* (1990). Devegowda *et al.* (1994) observed a significant reduction in the size of the Bursa of Fabricus in broilers fed diets containing 0.5 ppm AF. Viridi (1989) observed thymic aplasia and up to 30% reduced weight of bursae in chickens fed AF contaminated diets. The reduction in size of the thymus is believed to reduce the production of the T-cells which control the cell-mediated immune responses including delayed hypersensitivity. The decreased sensitivity reaction is therefore probably directly related to the reduction of the thymus (Table 2). The reduction in the size of the bursa probably affects production of the B type lymphocytes. However, this was not reflected in the reduction in the percentage of circulating lymphocytes in this trial. The reduction in the circulating heterophils and together with the differential reduction in bursae and thymic lobule weights indicates that AF had a

greater effect on the thymus and cell-mediated immunity than on the bursa and humoral immunity. This differential effect of AFB1 on the immune system of animals has been observed before (Pier and McLoughlin, 1985). The duration of tonic immobility also decreased significantly when birds were fed AFB1 in their diet (Table 1). TI duration is used as a measure of fear and as an indicator of severity of reaction to stress. Birds that respond more to stress have been found to have longer TIs. AFB1 decreases the ability of the bird to react to stress thus making it more susceptible to the effects of antigens such as pathogenic agents. In particular AFB1 decreases the ability of birds to react to pathogens that stimulate cellular immunity such as coccidiosis, Infectious Laryngo Tracheitis and Marek's disease. Birds with AFB1 in their diets are therefore more likely to die of these infections. If tonic immobility has been developed as a protective mechanism to save birds caught by dogs and foxes then AFB1 consumption makes birds more susceptible to predation by decreasing their tonic immobility, thereby making them more attractive targets to predators.

IV. CONCLUSIONS

Aflatoxin B1 at a dietary level of 0.9 ppm has a wide range of effects. It causes decreased growth and feed intake and decreased cellular immunity (H/L ratio, delayed skin hypersensitivity), relative size of thymic lobules and Bursa of Fabricus and tonic immobility. All these effects mean that birds consuming AFB1 are highly susceptible to disease agents and stress.

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TRI-PHOSPHOR FLUORESCENT LIGHTING FOR BROILERS - A PILOT STUDY

P.C. GLATZ¹, T. COVERDALE², M. PARTINGTON², CALVIN EICHNER³
and COREY EICHNER³

Summary

A field trial was conducted in two identical commercial broiler sheds on the same property and compared the behaviour of broilers receiving tri-phosphor fluorescent lighting in one shed with those receiving cool-white fluorescent lighting in another shed. There were no differences in the active behaviours of birds provided with the tri-phosphor lighting including the number of birds feeding and drinking. It is suggested that one tri-phosphor tube could be used to replace double cool-white light fluorescents in broiler sheds. While the initial purchase costs of tri-phosphor tubes are higher, their efficiency is superior to conventional cool-white tubes and power usage is lower.

I. INTRODUCTION

The two main types of artificial light namely, incandescent and fluorescent are presently in use on broiler farms. The incandescent radiant source is a hot filament of tungsten but tends to be deficient in the blue end of the spectrum, contains virtually no ultraviolet light, emits much of its light output as yellow and red, and releases its maximum energy as infrared radiation. Unlike incandescents, fluorescent lamps generate visible light by non thermal mechanisms. Fluorescent sources can produce different kinds of light depending on the phosphors (fluorescent substances) they contain. Typically, however they produce a rather distorted spectrum of light, which contains a limited portion of the total spectrum. The commonly used cool-white fluorescent light produces proportionally less blue and purple light than some other fluorescent tubes.

The influence of photoperiod, light intensity, colour, intermittent lighting and ahemeral cycles on husbandry, behaviour, performance and health of poultry has been reviewed by Kirkden *et al.*, 1995; Gordon, 1994; Morris, 1994. Little attention, however, has been given to studies comparing the effects of type of fluorescent lighting on bird behaviour and performance. In humans Hollwich and Dieckues (1980) found that cool-white fluorescent light stimulated an increase in stress (higher levels of ACTH and cortisol) compared with light from a tube simulating sunlight. This may explain the increase in fidgety behaviour and fatigue reported by patients exposed to cool-white fluorescent lighting. Phototherapy using fluorescent tubes simulating day light is increasingly being used by practitioners to treat a range of stress conditions in humans including depression (Kripke, 1985).

In the Poultry industry there has been no scientific evaluation or demonstration of tri-phosphor lights under commercial conditions in Australia, which has slowed the introduction of this new technology. The following field trial was conducted to compare the behaviour of meat birds provided with tri-phosphor fluorescent lighting versus conventional cool-white fluorescent light.

¹ SARDI, Pig and Poultry Production Institute, University of Adelaide, Roseworthy, SA 5371.

² Nelson Lamps Aust Pty Ltd, 37-39 Kolara Road, West Heidelberg, Vic 3081 and 22 George St, Stepney, SA 5069.

³ Windsor Poultry Farm, Windsor, SA 5501.

II. METHODS

(a) Sheds

This field trial was conducted in two identical commercial broiler sheds on the same property and compared behaviour of broilers provided with tri-phosphor fluorescent lighting in one shed and cool-white fluorescent lighting in another shed. Each shed was 100 m by 12 m and housed 20,000 broilers. The ridge vent in the roof and the side panels on the shed were closed during the period of the experiment in winter. Temperature control was similar in each shed. Ventilation for birds was provided by four fans at both ends of the shed drawing air through pads located in the middle of the shed. Natural light entered at the ends of the shed when the fans operated.

(b) Lighting

Lighting comprised 13 double cool-white 40 w fluorescents fitted in the centre of one shed. In the other shed, cool-white light fluorescents were removed and 13 single 37 w tri-phosphor fluorescent tubes installed. Light intensity was measured using a Gossen Panlux Light Meter at bird level near outer wall and directly under the light source. Lighting was continuous in each shed from 0-6 weeks.

(c) Behavioural measurements

The activities of birds were scanned using the instantaneous sampling procedure of Lehner (1996). Each shed had three feeder lines and four nipple lines. At three weeks of age the number of birds feeding from 30 feed pans and drinking from 40 nipple drinkers were counted within the vicinity of each of 11 lights. End lights were not included. Behaviour was scored for 80 birds under each light in each shed. Only the bird located midway between the side wall and nipple line or between the nipple lines and feed lines was scored. The percentage of birds sitting, standing, walking or engaging in other activities was determined. Other behavioural activities recorded were floor pecking, feather pecking, preening, dustbathing, sparring, fighting, feather ruffling, wing flapping and head scratching. All behaviour observations were made between 0900 and 1700 h. The process involved instantaneous sampling of behaviour of birds under a light in one shed (20 min) followed by observations in the other shed. Initially when the observer entered the shed, birds were disturbed and took about 10 min to settle. The observer moved back and forward between the sheds until behaviour of birds under 11 lights in each shed had been scored. Data obtained on birds in the vicinity of each light was considered to be a replicate for the purposes of analysing the results. The experiment was analysed to determine the effect of light quality on behaviour of broilers using the general linear models procedure using SAS GLM (Statistical Analysis Systems Institute Inc., 1988).

III. RESULTS AND DISCUSSION

The light intensity from the tri-phosphor tubes at floor level directly under the light source was 89 lux which was significantly ($P < 0.05$) greater than intensity (68 lux) of two equivalent wattage cool-white light fluorescents. On the sides of the shed where the birds generally rested, light intensity was similar for both light sources (9.5 lux and 8.3 lux for tri-phosphor and cool-white lights respectively). Given the higher light intensity from the tri-

phosphor tubes it was expected that more birds would be walking or engaged in other active behaviours. This was not observed. There were no differences between the light sources in the percentage of birds walking (Table 1) or in the number of birds feeding and drinking (Table 2). In support of these observations, staff caring for the birds said they could not detect any obvious differences in bird activity or any other behaviours. Likewise they reported there were no differences in broiler feed intake, feed conversion and mortality that could be attributed to light source. The owner manager suggested that if he was installing the tri-phosphor lights in sheds he would space the lights further apart because of their higher light intensity, even though tri-phosphor tubes had similar operating wattage to the cool-white tubes.

Table 1. Effect of tri-phosphor and cool-white light fluorescent lighting on proportion of birds sitting, standing, walking or engaged in other activities.

Treatment	Sitting (%)	Standing (%)	Walking (%)	Other behaviour (%)
Tri-phosphor	48.3	18.9	23.7	9.1
Cool-white	44.2	21.2	25.6	8.6
P	NS	NS	NS	NS

NS = differences are not significant ($P > 0.05$)

Table 2. Effect of tri-phosphor and cool-white light fluorescent lighting on number of birds feeding and drinking.

Treatment	Light Intensity (lux) near wall	Light Intensity (lux) under light	No. Birds feeding per hopper	No. Birds drinking per nipple
Tri-phosphor	9.5	89.2	6.3	1.5
Cool-white	8.3	68.3	6.1	1.5
P	NS	0.02	NS	NS

P = probability. NS = not significant in analyses of variance ($P > 0.05$).

This pilot study has provided some evidence that tri-phosphor lighting does not cause major changes in behaviour of 3 week-old broilers. Before firm recommendations can be made more detailed growth, behaviour and health studies are required over the growing period of the bird. It would appear that utilisation of this new technology lighting for poultry offers considerable savings in running costs. Data in table 3 presents the power costs of the continuous operation of one tri-phosphor lamp compared to two cool-white light fluorescents. Calculations are based on 12c/KiloWattHour (KWH) for peak usage (14h/day), 6.1c/KWH (10h/week day and all weekend). Power costs for one tri-phosphor tube is \$35/year compared to \$76/year for two conventional cool-white tubes. Further studies are required with a variety of alternative lights that are now available on the market so that sound guidelines can be provided to industry on distribution of lights in sheds and any possible effects on performance, behaviour and health.

In the interim, it is suggested that one tri-phosphor tube could be used to replace a double cool-white light fluorescent in broiler sheds. While the initial purchase costs of tri-phosphor tubes are higher, their efficiency is superior to conventional cool-white tubes and power usage is lower.

Table 3. Power costs for one tri-phosphor and two cool-white fluorescent tubes

	Lamp Type	
	Tri-phosphor fluorescent	Cool-white fluorescent
Operating wattage	37	40
Ballast loss	10	11
Total wattage (W) consumed	47	51
Number of lamps (L)	1	2
Peak consumption rate	12c/KWH	12c/KWH
Peak consumption h/day	14	14
Cost of peak consumption/day = ((Wx14)/1000) x 12 x L (c)	7.896 c	17.136 c
Off peak consumption rate	6.1c/KWH	6.1c/KWH
Off peak consumption h/day	10	10
Cost of off peak consumption/day = ((Wx10)/1000) x 6.1 x L (c)	2.867 c	6.222 c
Total daily cost/week day (c)	10.763 c	23.358
Total week day cost (c)	53.815 c	116.69 c
Cost of weekend consumption = ((Wx48)/1000) x 6.1 x L (c)	13.761	29.866
Total cost/week (c)	67.6 c	146.6 c
Annual cost	\$35.15	\$76.23

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DATA OBTAINED FROM AUTOMATIC WEIGHING IN COMMERCIAL BROILER FLOCKS

S. DUAN-YAI¹, B.A. YOUNG¹, J.A. COUTTS² and J.B. GAUGHAN¹

Summary

Data were collected from 4 commercial broiler flocks (Steggles Poultry Ltd.) to evaluate the uniformity of broiler body weight over time. Broiler weights were collected using an automatic device, Computrol Mini Weight System (PEC Co. Ltd.). Number of weights recorded, uniformity, SD and CV were found to be different over time ($P < 0.01$). Number of weights recorded daily peaked at week 1 and then gradually decreased as the broilers grew. The highest uniformity in broiler weights was obtained in the spring flock.

I. INTRODUCTION

Body weight and distribution of body weight in broiler flocks are indicative of flock quality and management status. In any commercial flock, regular manual weighting is time consuming and stressful to the broilers. An alternative approach is the use of automatic weighing in which body weight and uniformity can be checked daily. The objective of this paper is to illustrate the mean growth pattern and distribution of body weights over time in commercial broiler flocks, as a potential support for daily management practices.

II. MATERIALS AND METHODS

Data were collected from 4 flocks, winter 1996, spring 1996, summer 1997, and autumn 1997 at The University of Queensland (Gatton Campus). Each flock comprised 5000 broilers (Steggles Poultry Ltd.) fed *ad libitum* on commercial starter diet for the first 3 weeks and grower diet for the last 2 weeks. The chickens were restricted to 1/6, 1/4, and 1/2 of the floor area for the first, second, and third week of the brooding period respectively before being given access to the full house in weeks 4 and 5 at a density of 14/m². The lighting regime was 24 hour through the test period.

The automatic weighing device, Computrol Mini Weight System (PEC Co. Ltd.), included a platform scale linked to a computer and a printer. The scale was placed between one feed line and drinkers with a 30 m long cord connected to the recording computer kept separately in a dust-free area. The scale was calibrated to a 1 kg standard weight. The weighing range was 0-5 kg within certain limits of daily standard growth. Day-old broilers were induced to stand on the scale by spreading some feed on the weighing platform. Information was provided diurnally including date, number of weights recorded, individual weight, mean weight, and uniformity (the percentage of the flock within 10% of the average weight at any time). In this study, data obtained in the morning period were selected as being representative for that day.

¹ Department of Animal Production, The University of Queensland, Gatton, Qld 4345.

² Rural Extension Centre, The University of Queensland, Gatton, Qld 4345.

III. RESULTS AND DISCUSSION

The number of weights recorded, uniformity, SD and CV were all found to be significantly affected by week of the growing period (Table 1). The only significant difference among flocks raised in different seasons was uniformity. The spring flock showed a higher uniformity compared to summer, autumn and winter (70 vs. 65, 62 and 62%).

Table 1. Data obtained from automatic weighing of four broiler flocks.

Data	wk 0	wk 1	wk 2	wk 3	wk 4	wk 5
No.weights (birds)	113 ^b	619 ^a	583 ^a	366 ^{ab}	319 ^{ab}	223 ^b
BW \pm SD (g)	43 \pm 4.2 ^a	140 \pm 14.7 ^b	393 \pm 42 ^c	703 \pm 81 ^d	1037 \pm 138 ^e	1508 \pm 169 ^f
CV (%)	9.7 ^b	10.6 ^{ab}	10.7 ^{ab}	11.5 ^{ab}	13.3 ^a	11.2 ^{ab}
<i>Uniformity (%)</i>						
Winter	69	58	65	68	56	57
Spring	81	61	69	68	70	73
Summer	80	59	64	62	57	65
Autumn	66	59	62	58	60	66

Means in the same row with different superscripts are significantly different ($P < 0.01$).

The number of weights recorded depended on the number of broilers standing on the scale. These numbers were found smallest at day 1, peaked at day 7 and then gradually decreased (Figure 1). It was observed that day-old broilers took a couple of hours getting used to the scale and its height (60 cm) while the heavier broilers of weeks 4 to 5 spent most of the time less active and on the litter floor. However, the number of records in this study exceeded 1% of the flock, which according to Fattori *et al.* (1994) and Turner *et al.* (1984) is sufficient to obtain a reliable sample of body weights.

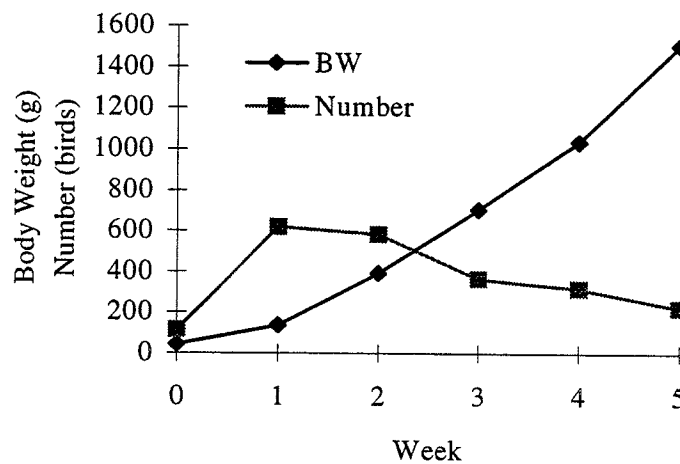


Figure 1. Body weight and number of weights recorded in 4 broiler flocks.

While SD of body weights increased over time, CV was relatively constant (Figure 2). Excluding the value on day 1 which was 9.7%, the CV range during the growing period was only 3% (10.6 - 13.3). These values could be used as a criterion to compare the similar flocks. A higher value may indicate an unusual situation. Experience from a similar flock, due to a leak from a water pipe, showed CV as high as 17%.

However, the range of CV could be very different among different flocks. The value of the growing period (week 1-5) from Doyle and Leeson (1989) was 6.2 to 14.5% while data of Newberry *et al.* (1985) started with 16.6% at week 1 and steadily decreased to 9.8% at week 5. Therefore caution must be taken when using CV to evaluate between flock performance. The CV range from this study was particularly reliable for a flock of 5000 Steggles unsexed broilers, at a density of 14/m² and *ad libitum* fed. Further experiments on different management strategies such as feed restriction or sexes reared separately could be undertaken to determine the effect on CV range.

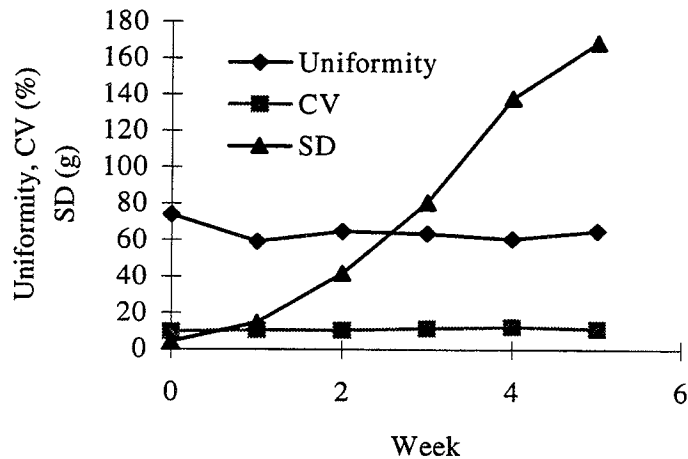


Figure 2. Uniformity, CV and SD of body weight from 4 broiler flocks

Although SD was not a constant value, it could be used to classify body weight range of a flock. According to the normal distribution, 68% of the flock would be expected to have a body weight within mean \pm SD, 95% of the flock would be expected to have a body weight within mean \pm 2SD, and 99% of the flock would be expected to have a body weight within mean \pm 3SD. This information could be used to support management decisions especially at market age. Table 2 is the example calculated from the present study.

Table 2. Body weight interval of broilers at week 5.

Proportion within flock	Body Weight Range (g)
68%	1369-1647 (mean \pm SD)
95%	1230-1786 (mean \pm 2SD)
99%	1091-1925 (mean \pm 3SD)

IV. CONCLUSION

Automatic weighing units can provide a sufficient number of weight records to obtain a reliable estimate of the mean and variance of flock body weight. SD was closely correlated to mean body weight in this study, which resulted in relatively constant CVs during the growing period. It is suggested that variation in CV could be used to evaluate possible disruptions to flock performance, although caution must be applied when using CV as the criterion, as different flocks may have their own specific range.

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OBJECTIVE MEASUREMENTS OF THE QUALITY OF EMU LEATHER

R.A. COLEMAN and J.G. DINGLE

Summary

The size of the tanned skin from six emus from the one farm was measured. Samples of leather from six locations on each side of the six emu skins were measured for thickness, ability to stretch over a shoe toe (lastometer), tear resistance, softness and flexibility. There was no deterioration of the tanned, dyed and finished leather after flexing 10000 times. There was no significant difference between the left and right sides of the skin for any of the readings with the exception of ability to stretch over a shoe toe which showed that samples from the left side failed the lastometer test more often than samples from the right side. There were significant differences between the skins in thickness, lastometer, tear strength and softness measurements. Thickness was related to size of the skin. There were significant differences between the samples from different locations in thickness, ability to stretch over a shoe toe, tear strength and softness. The values recorded indicated that emu leather is suited to apparel and accessory manufacture.

I. INTRODUCTION

Leather is a complex mixture of connective tissue fibres oriented in different directions and held together by a number of cross-links (Sharphouse, 1995). The number of connective tissue fibres, their strength, orientation and bonding may vary depending on the breed, age, sex, nutrition, environment and experience of the bird (Tancous, 1992). Therefore the quality of emu skins is likely to vary from one individual to another. In addition, during the time of flaying, mechanical damage may occur, and later damage may occur due to conditions of curing and storing which may result in loss in strength or grain character. In general, leather is sold on the basis of size (area), thickness, and visual appearance of the surface grain and incidence of faults visible to the naked eye.

To compare different samples of leather it is essential that quality be defined in objective terms. These measurements will also assist buyers and sellers to determine the suitability of a particular skin for a particular purpose. Emu leather is used for making apparel, high fashion shoes, and accessories such as gloves, wallets, handbags, and key cases. This paper reports on the use of six objective measurements of emu leather to determine if it is most suited to its present uses.

II. MATERIALS and METHODS

(a) Emu skins

Six skins were obtained from a batch of 50 emus of mixed age (10 months to 3 years) and sex and processed at the Cherbourg abattoir. All birds had been fed a grain-based diet and the average carcass weight for all the emus was 17.0 kg.

(b) Tanning

The six skins were washed and placed in an ice chilled brine solution immediately after flaying. After 24 hours in the brine solution, they were taken to the tannery where they underwent liming, pickling, and tanning using chromium sulphate at 7% of wet tannage. This produced a soft leather, pale blue in colour, which was then dyed and finished in black. The flesh surface of the body, but not the neck portion, of the skin was trimmed by buffing..

(c) Objective tests

Surface area (yield). Surface area was measured in square decimeters using a purpose-built machine which registers when rollers move over the surface of the skin. The distance the rollers travel was calculated and a surface yield was recorded and stamped on the skin. In the other five objective tests, six locations were measured on each side of each skin. The locations were: (1) butt; (2) leg; (3) centre back; (4) chest; (5) wing area; (6) neck. These locations were chosen to provide information about the variations in skin quality across the skin.

Thickness. A simple micrometer gauge was used to measure thickness between the upper (grain) surface and the lower (flesh) surface.

Tear strength. A standard rectangle 50 mm x 25 mm was cut out of each location, with a slot 20mm x 5 mm in the centre by the use of a heavy press and a standard metal die. The rectangle was oriented with the long axis parallel to the backbone of the bird. A load of 10 kg was applied to two jaws placed on each side of the slot and moved apart at a speed of $100 \pm 20 \text{ mm min}^{-1}$. The tear load at the time the leather strip tears was measured in kilogram force (kgf) units.

Lastometer test. A 7mm long rod applied at 20 kg pressure was pushed into the flesh side of the leather. If it broke the grain surface the result was recorded as a failure and was given a value of 2. If it failed to break through the grain surface the sample was given a value of 1.

Softness test. A solid cylinder of metal was lowered into a hollow cylinder of metal with the leather sample stretched between the cylinders. The distance the solid cylinder moves into the hollow cylinder is a measure of the softness of the leather.

Flexibility test. A piece of leather 650mm x 650 mm was cut from the skin at each location by use of a press and metal die. This piece of leather was attached to an oscillating machine which flexed the leather without tensioning it. The machine was set to make 10000 oscillations/ sample and the grain surface was then examined under a microscope and any crack in the surface was recorded as a failure.

The experiment was a 6 x 6 x 2 factorial design with the factors being skins x locations x sides. Factorial analysis was undertaken using SAS to calculate the analysis of variance. Also the differences between means were tested for significance using Duncan's multiple range test at the 5% level of probability.

III. RESULTS AND DISCUSSION

The mean results of each measurement for the six skins are shown in Table 1(a) and for the six locations are shown in Table 1b. As there was no failure in the flexibility test, these results are not shown. There was no significant difference in mean readings between the two sides except for the lastometer test, where the left side samples failed significantly

more often than the right side samples. There was no obvious reason why the left side was weaker than the right.

Table 1. Mean objective measurement of emu skins¹.

(a) Skins						
Skin number	Size (dm ²)	Thickness (mm)	Lastometer	Tear strength (kgf at 10 kg)	Softness (0.01 mm)	
1	69	0.50 ^{ab}	1.17 ^{ab}	3.00 ^b	54.92 ^a	
2	71	0.60 ^a	1.00 ^b	3.83 ^a	50.92 ^b	
3	66	0.49 ^b	1.25 ^{ab}	3.12 ^b	49.59 ^{ab}	
4	70	0.52 ^{ab}	1.33 ^a	3.67 ^{ab}	48.42 ^{bc}	
5	87	0.60 ^a	1.25 ^{ab}	3.29 ^{ab}	47.25 ^{bc}	
6	70	0.51 ^{ab}	1.00 ^b	3.29 ^{ab}	46.08 ^c	
Treatment means		72	0.54	1.17	3.37	49.53

(b) Locations				
Locations	Thickness (mm)	Lastometer	Tear strength (kgf at 10 kg)	Softness (0.01 mm)
1 butt	0.53 ^b	1.08 ^b	3.25 ^c	45.25 ^c
2 leg	0.48 ^b	1.08 ^b	2.54 ^d	49.75 ^b
3 centre back	0.62 ^a	1.17 ^{ab}	4.29 ^a	45.25 ^c
4 chest	0.53 ^b	1.17 ^{ab}	3.58 ^{bc}	51.75 ^{ab}
5 under wing	0.45 ^b	1.42 ^a	2.62 ^d	50.83 ^{ab}
6 neck	0.62 ^a	1.08 ^b	3.92 ^{ab}	54.33 ^a

¹ Means without the same superscript are significantly different (P<0.05).

The skin areas varied from 66 to 87 dm². All the skins included a full neck. The skin size is a reflection of bird size. American research reported that the average tanned emu skin's surface area was 50 to 65 dm² (Kateswara and Wm, 1996). The report indicated difficulty in retaining the neck region in commercial tanning operations. During the last year, the Cherbourg tannery developed a tanning method, which ensured that the whole neck could be tanned without the knotting which caused loss of the neck.

The leather thickness varied from 0.49 mm to 0.60 mm for the different skins, being significant. The largest skins were also the thickest so possibly skin increases in thickness with age of bird. The range of thickness was greater for the different locations than for the different birds. The thickest skin was in the centre back and neck region. Skin samples from these locations were significantly thicker than skin samples from the other four locations. The average thickness of the USA skins measured by Kateswara and Wm (1996) was between 0.49 mm to 0.60 mm. The thinness of our skins might be due to the low level of nutrition available to the birds.

There was a significant difference between skins and between locations on the skin for the lastometer test. Grain failure did not appear to be related to skin size because two large skins had the highest and the lowest readings. Two skins had no grain failures, indicating that these skins were quite suitable for shoe leather as they could be stretched over a toe cap without breaking. There were significant differences between locations on the skin. Samples from the wing area were significantly weaker and "popped" more frequently in the lastometer test than samples from the butt, leg and neck area.

The tear strength of the skins differed with skin size, the largest skin being significantly resistant to tearing than the smallest two skins. Tear strength also varied across the skin, with the greatest strength being in the centre back and neck areas, which were also the thickest parts of the skin. Skin from the centre back was significantly stronger than skin from any other location and skin from the neck was significantly stronger than skin from the butt, leg or wing area.

Skins also differed significantly in softness but softness did not appear to be related to skin thickness, tear strength or "lasting" ability. Skin from the neck was significantly softer than skin from the leg which was significantly softer than skin from the butt and centre back.

IV. CONCLUSIONS

Emu leather from different individuals and from different locations on the one individual varies in size, thickness, lasting ability, tear strength and softness. The factors causing these differences have not all been identified, but some of the differences are related to size, which is probably related to age and nutrition of the bird. More work is needed to determine how sex, nutrition and handling affect leather quality. However the leather is suited to its present uses for apparel and accessories.

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REPRODUCTIVE PERFORMANCE IN JAPANESE QUAIL SELECTED FOR BREAST MEAT YIELD

B. POPOVIC and R.A.E. PYM

Summary

Measures were made of correlated response in reproductive performance in lines of Japanese quail (*Coturnix coturnix japonica*) selected over eight generations for increased breast weight (Line BWI), increased liveweight (Line LWI), or increased or decreased breast proportion (Lines BPI and BPD respectively). Egg production and hatchability were depressed and the onset of sexual maturity delayed in lines BPI, LWI and BPD. Egg weight increased in Line BWI but was unchanged in the three other lines. The study indicated that reproductive performance in Japanese quail was generally depressed by selection for increased liveweight and/or breast meat weight or proportion.

I. INTRODUCTION

Selection for increased growth rate in avian species typically results in depressed egg production, fertility and hatchability (e.g. Marks, 1979; Siegel and Dunnington, 1985; Pym 1985; Bunan and Pym, 1991). While reproductive performance is generally reduced by selection for aspects of body composition or food utilisation efficiency, the reduction is considerably less than from such selection when combined with selection for increased growth rate (Bunan and Pym, 1991; Pym 1985). The negative effects on reproductive performance of like selected lines are countered to a degree, but not completely, by appropriate nutritional and lighting management of the selected stock, as applied to broiler breeders in commercial practice.

It is of prime importance to breeders to have information on the likely consequences on reproductive performance of selection for the different performance traits in juvenile birds. Because of the high relative value of breast meat in poultry species, we commenced a selection experiment in 1993 for aspects of breast meat yield in Japanese quail, using real-time ultrasound, and reported at this meeting last year on direct and correlated response in growth, breast meat yield, feed efficiency and body composition to five generations of selection (Popovic and Pym, 1997). This paper reports on a study of reproductive performance in the lines to eight generations of selection. We are unaware of any reports of correlated response in reproductive performance to selection for breast meat yield in poultry.

II. THE SELECTION EXPERIMENT

From a population of Japanese quail (*Coturnix coturnix japonica*) previously selected for increased 42d liveweight for about ten generations, five lines were generated with subsequent selection for either; increased 42d liveweight (Line LWI), increased breast meat yield (g, Line BWI), increased breast meat proportion (g/kg, Line BPI), decreased breast meat proportion (g/kg, Line BPD), or at random (Line C). Real-time ultrasound was the method used to measure breast muscle depth. At each generation each line was reproduced by mating each of 12 males to three females, and eggs were saved for four weekly hatches.

Division of Farm Animal Studies, The University of Queensland, St Lucia, Qld 4072.

The hens were each placed in a pair-breeding cage and the males were moved daily between the three females, spending 24h every 72h with each female. Records were taken of egg production and hatchability for each of the 36 females in each line across the four hatches.

Females sampled at random from two hatches of birds from the eighth selected generation of each line were reared to six weeks of age, when 36 birds per line were allocated as above to the breeding cages. Records were taken on each hen of liveweight on placement in the cages at 42d, age at first egg and egg production to 11 weeks of age. All eggs produced over this period were weighed each week to determine correlated response in egg weight in the four selected lines.

III. CORRELATED RESPONSES TO SELECTION

The correlated responses in egg production in the four lines to seven generations of selection are shown in Figure 1. Response in each generation was measured relative to performance in the control line. There was a general trend in all lines, with the exception of the low breast proportion (BPD) line, for production to decrease as selection proceeded, but the trend was decidedly more pronounced in the LWI and BWI lines than in the high breast proportion (BPI) line. It is significant to note that liveweight gain was substantially increased in the former two lines but unchanged in the two breast proportion lines (Pym *et al.*, 1998).

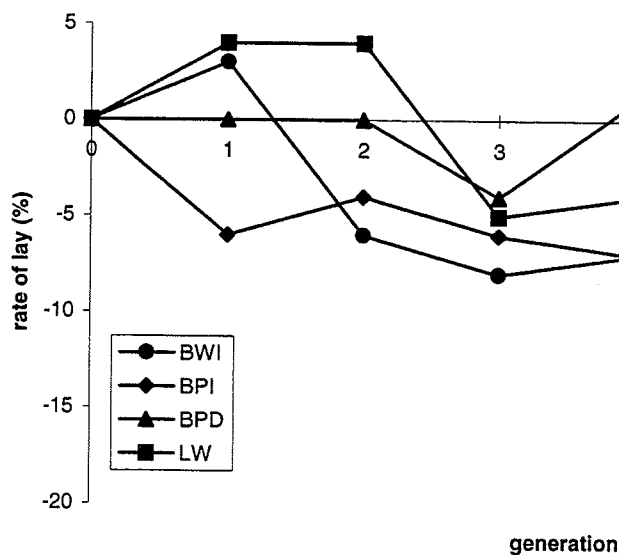


Figure 1. Correlated response in rate of lay (%) in hens from the four selected lines over 7 generations of selection.

The correlated responses in hatchability to seven generations of selection in the four lines are shown in Figure 2. There was a general trend in all lines for hatchability to decrease with selection, although the reduction in hatchability was considerably less in the low breast proportion (BPD) line than in the three other lines.

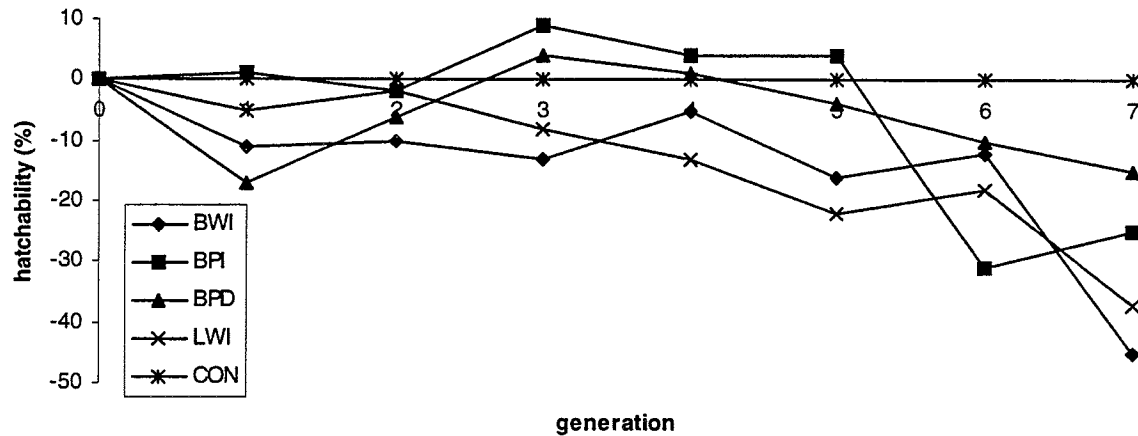


Figure 2. Correlated response in hatchability (%) in the four selected lines over 7 generations of selection.

Mean 42d liveweight, age at first egg, egg production to 11 weeks and egg weight from hens in the five lines at generation 8 are shown in Table 1.

Table 1. Mean 42d bodyweight (g), age at first egg (d), egg production to 11 weeks of age (eggs/bird) and egg weight (g) in the five lines after 8 generations of selection.

Line	42d body weight	Age at first egg	Egg production	Egg weight
BWI	232 ^a	50.4 ^a	10.7 ^c	11.8 ^a
BPI	211 ^b	49.7 ^a	11.2 ^c	10.6 ^b
BPD	205 ^b	46.6 ^b	14.7 ^a	10.7 ^b
LWI	235 ^a	49.9 ^a	11.0 ^c	10.7 ^b
C	209 ^b	47.4 ^b	12.8 ^b	10.3 ^b
LSD _{0.05}	8	2.3	1.5	0.5

The BWI and LWI birds were heavier than birds in the three other lines ($P < 0.05$) and age at sexual maturity was delayed in the BWI, BPI and LWI lines ($P < 0.05$), but unaffected in the low breast proportion (BPD) line. Egg production was similar and lowest in the BWI, BPI and LWI lines, intermediate in the unselected control (C) line, and highest in the low breast proportion (BPD) line. Egg weight was significantly higher in the high breast weight (BWI) line than in the four other lines.

IV. DISCUSSION

The negative correlated response in egg production and hatchability in the four lines (Figures 1 and 2), is in keeping with the findings of previous studies where selection has been exercised for increased liveweight gain and aspects of body composition (Marks, 1979; Siegel and Dunnington, 1985; Pym, 1985; Bunan and Pym, 1991) The more pronounced depression in reproductive performance in the increased liveweight (LWI) and breast weight (BWI) lines is also in keeping with the findings of Bunan and Pym (1991) where lines of chickens selected for increased liveweight alone or in combination with reduced fatness had considerably lower reproductive performance than lines selected for either high or low body fat alone, where there was no correlated response in growth rate.

In the present study, there was a substantial correlated response in growth rate in both the LWI and BWI lines but no such correlated response in the high and low breast proportion lines. It would appear that increased juvenile body weight as a consequence of selection is a prime factor in eliciting a negative response in reproductive performance.

The delay in the onset of sexual maturity in the increased liveweight, breast weight and breast proportion lines at generation eight is in keeping with the earlier study of Pym (1985) with chickens, where the control line came into production some 10 days before the lines selected for increased growth rate or appetite; the lean feed efficiency line in that study was delayed by a further 12 days. In the present study, the low breast proportion line, which had a high proportion of body fat (Pym *et al.*, 1998), came into lay even earlier than the control birds. Body composition would appear to be an important determinant of the onset of sexual maturity both between and within genotypes. In the latter case a certain level of body fatness, characteristic of that genotype, must be achieved before the bird will commence lay (Bunan, 1990).

The positive correlated response in egg production to 11 weeks of age in the BPD line at generation eight, is possibly a reflection of the earlier onset of lay in this line, but also of typical generation-to-generation variation in a breeding program. As shown in Figure 1, there is, however, a different pattern of correlated response in egg production in this line compared to the other selected lines, possibly associated with its reduced body protein deposition and/or higher fat deposition. It is somewhat unfortunate, but not unexpected, that this line with the most undesirable juvenile growth performance had the best reproductive performance. It is likely that reproductive performance of all lines could be improved by feeding and lighting techniques appropriate to the different genotypes (Bunan, 1990; Bunan and Pym, 1991).

The significance of the increased egg weight at generation eight in the increased breast weight (BWI) line, which was not observed in the high liveweight (LWI) line, may be explained by differences in protein utilisation and deposition in the lines. It would be expected that these two lines, which were heavier than the other lines, would also lay larger eggs. The greater amounts of protein deposited as muscle in the breast of the BWI birds during early growth may reflect a similar capacity to deposit protein in the egg. This was, however, not observed by Bunan and Pym (1991) where the high liveweight and high lean liveweight lines of chickens produced eggs of similar weight.

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IDEAL AMINO ACID PROFILE FOR BROILER CHICKENS FROM 20 TO 40 DAYS OF AGE

S. MACK¹, D. BERCOVICI², G. DE GROOTE³, B. LECLERCQ⁴, M. PACK⁵,
J.B. SCHUTTE⁶ and S. VAN CAUWENBERGHE²

Summary

A series of experiments was conducted to determine the ideal ratio of lysine (Lys) to methionine (Met), threonine (Thr), tryptophan (Trp), arginine (Arg), valine (Val) and isoleucine (Ile) and to assess the adequate dietary lysine concentration for optimum performance in broiler chickens of two commercial strains during 20 to 40 days of age. One identical basal diet was used in all experiments. It mainly consisted of maize and soyabean meal and contained 172 g crude protein and 3158 kcal AMEn per kg. For each experiment the basal diet was adequately supplemented with all essential amino acids except the one to be tested which was supplemented in six graded levels in exchange with maize starch. One (Met, Trp, Arg, Val, Ile) or two (Lys, Thr) feeding trials were conducted per tested amino acid and the response in weight gain, feed/gain and breast meat yield examined. By subjecting the data to broken line regression analysis, the ideal amino acid ratio relative to Lys was calculated to be: 75 % Met+Cys, 63 % Thr, 19 % Trp, 112 % Arg, 71 % Ile and 81 % Val. Concentration for optimum feed/gain was calculated to be 12.2 g/kg total or 11.5 g/kg true excreta digestible lysine by the application of exponential regression analysis.

I. INTRODUCTION

Amino acid (AA) requirements of livestock are influenced by a multitude of dietary, environmental and genetic factors. Thus it is almost impossible to address all potential combinations of factors with dose-response experiments looking into the full range of essential AA. Swine nutritionists were the first to solve this problem by expressing AA requirements as ratios to lysine (Wang and Fuller, 1989; Chung and Baker, 1992). This method is based on the premise that although the AA requirements change due to the above mentioned factors, the ideal ratio of essential AA to lysine would remain unaffected. Once the ideal ratio of essential AA to lysine is established, one can concentrate on determining the lysine requirement under a variety of conditions and can calculate the requirements for other indispensable AA by applying their ideal ratio to lysine. Lysine was chosen as the reference amino acid in pigs and poultry because it is almost exclusively used for protein accretion after absorption and it is relatively easy to analyse in feedstuffs. Furthermore, there is a large body of data available on the response in performance to different lysine levels under varying conditions. Ideal amino acid ratios should be based on digestible AA. This is especially important when feed ingredients other than maize and soyabean meal are included in diet formulations. For many years there has been ongoing research on optimal AA ratios for broiler chicks.

¹ Degussa AG, Feed Additives Div. Applied Technol., PO Box 1345, 63403 Hanau Germany.

² Eurolysine, 153 rue de Courcelles, 75817 Paris Cedex 17, France.

³ Rijksstation voor Kleinveeteelt, Burg. Van Gansberghelaan, 92, 9820 Merelbeke, Belgium.

⁴ INRA, Station de Recherches Avicoles, 37380 Nouzilly, France.

⁵ Finnfeeds International, Market House, Marlborough, Wiltshire, SN8 1AA, UK.

⁶ TNO-ILOB, PO Box 15, 6700 AA Wageningen, The Netherlands.

However, most of the work has been done with young birds from hatching to about three weeks of age, whereas the database for broilers from three to six weeks of age is still rather limited (Hurwitz *et al.*, 1978; Baker, 1994). The present study investigated the response of broiler chickens from 20 to 40 days of age to graded dietary levels of lysine (Lys), methionine (Met), threonine (Thr), tryptophan (Trp), arginine (Arg), valine (Val) and isoleucine (Ile). The aims were to derive an ideal ratio for the tested amino acids and to determine the dietary lysine concentration for optimum bird performance.

II. MATERIALS AND METHODS

Nine dose-response experiments were conducted at three European research institutes (INRA Station de Recherches Avicoles, Nouzilly, France; Rijksstation voor Kleinveeteelt, Merelbeke, Belgium; TNO-ILOB Institute, Wageningen, The Netherlands). One (Met, Trp, Arg, Ile, Val) or two (Lys, Thr) experiments were conducted per AA each comprising a total of 960 to 1440 birds, six treatments and four to six replicates each with 32 to 48 birds. After termination of the experiment 40 or 48 birds per treatment were subjected to carcass analysis. The experiments utilised male birds of two commercial strains: ISA (Lys, Thr, Ile, Val) and Ross (Met, Lys, Thr, Trp, Arg). For the first 20 days posthatching the chicks were fed a commercial starter diet adequate in all nutrients. At 20 days of age they were weighed and assigned to floor pens following a randomized block design. Stocking density was 12 to 13 birds per square metre throughout all experiments. At each research site the birds were housed in insulated, artificially heated, ventilated and illuminated broiler units under similar environmental conditions with mean air temperature in the broiler units during the experimental period being adjusted to 20°C. Experimental diets and water were offered *ad libitum*. The basal diet consisted mainly of maize and soyabean meal and contained 13.2 MJ AMEn and 172 g CP per kg. For each tested AA true fecal digestibility (TFD) was determined (Lessire, 1990) except for Trp. Total and TFD AA content of the basal diet is given in Table 1.

Table 1 Total and TFD amino acid content of the basal diet.

	AA content in basal diet (g/kg)							
	Met	Met+Cys	Lys	Thr	Trp	Arg	Ile	Val
Total	2.4	5.1	7.2	5.5	1.5	8.9	6.1	6.8
TFD	2.3	4.5	6.5	4.8	1.3 ¹	8.4	5.6	6.1

¹ Assumed TFD coefficient of 0.88 = Table value for soyabean meal (Eurolysine, 1995)

For each experiment the basal diet was fortified with all AA under investigation, except the one to be tested, to contain 5.8 g Met, 8.5 g Met+Cys, 11.5 g Lys, 8.3 g Thr, 1.9 g Trp, 11.7 g Arg, 7.9 g Ile, and 9.2 g Val per kg of diet. In each experiment the amino acid to be tested was added to the basal diet in six graded levels (Table 2) in place of maize starch to form six experimental diets which were pelleted thereafter. Synthetic amino acids were assumed to be 100 % digestible. Experimental diets were formulated according to analysed crude protein and AA content of the individual feedstuff batches (Llames and Fontaine, 1994).

Weight gain, feed/gain, and breast meat yield data were subjected to one-way analysis of variance (ANOVA). Differences between treatment means were tested for significance ($P < 0.05$) using the Student-Newman-Keuls test (Snedecor and Cochran, 1980). Broken line

and exponential model were fitted to the data by means of a non-linear regression procedure (SAS Institute, 1987). For the exponential model the dose-related responses to dietary AA supplementation were modelled using the following equation (Schutte and Pack, 1995):

$$y = a + b(1 - e^{-c(x-d)})$$

where: y = weight gain or feed/gain or breast meat yield; a = intercept (performance accomplished with basal diet for the parameter investigated); b = maximum response to dietary concentration of the tested AA (= asymptote); c = curvature steepness; d = AA content of experimental diet I (g/kg); x = AA content of experimental diets II to VI (g/kg).

A modified Gauss-Newton iterative algorithm was employed to estimate the variables a , b , and c simultaneously. Tentative values for AA requirements were calculated at an arbitrarily chosen 95% of maximum response to amino acid supplementation.

Table 2 Tested AA supplementations to basal diet.

Experimental diet	Levels of AA added to basal diet (g/kg)						
	DL-Met	L-Lys	L-Thr	L-Trp	L-Arg	L-Ile	L-Val
I	0.9	0.8	0.0	0.0	0.0	0.0	0.0
II	1.5	1.7	0.6	0.1	0.9	0.5	0.5
III	2.1	2.6	1.2	0.2	1.8	1.0	1.1
IV	2.7	3.5	1.8	0.4	2.8	1.5	1.6
V	3.3	4.4	2.4	0.5	3.7	1.9	2.2
VI	3.9	5.3	3.0	0.6	4.6	2.4	2.7

III. RESULTS AND DISCUSSION

General performance level of the chicken was good and mortality rate was low (2-6%) in all experiments. Within each experiment no substantial differences in mortality occurred between treatments. Table 3 summarises treatment means for weight gain, feed/gain and breast meat yield (g or % of live weight).

The ideal amino acid ratio for weight gain calculated from broken line regression analysis and based on TFD amino acids was 100 (Lys), 75 (Met+Cys), 63 (Thr), 19 (Trp), 112 (Arg), 71 (Ile), 81 (Val). Break points of the broken line model applied to weight gain data were similar for both Lys experiments (8.5 g and 8.7 g TFD AA/kg diet for ISA and Ross birds, respectively) and identical in the Thr experiments (5.4 g TFD AA/kg diet). Ideal AA ratio was calculated from the mean value of the two Lys studies (8.6 g TFD Lys/kg diet). TFD for Trp was taken from the literature (Eurolysine, 1995). The calculated ideal AA ratio is well in line with results published by Baker (1994) except for Thr and Arg for which he reports 70% and 105% of the Lys requirement, respectively.

Accurate assessment of dietary lysine specifications is critical when applying the ideal amino acid profile for feed formulations because all other essential AA are related to it. The broken line model is not suited for that purpose. It neglects the biological law of diminishing returns and generally underestimates the optimum dietary AA content (Morris, 1989; Remmenga *et al.*, 1997). In contrast, the exponential regression model, that accounts for biological principles is sensitive enough to pick up small but economically important differences in performance at higher supplementation levels and allows for calculating the most economical lysine concentration (Pack and Schutte, 1995). Table 4 shows the Lys requirement estimated by exponential regression analysis at 95 % of the asymptotic response.

Table 3 Effects of dietary amino acid supplementation on broiler performance

AA	Strain	Parameter	Dietary treatment						SEM
			I	II	III	IV	V	VI	
Met	Ross	Weight gain (g)	1573 ^c	1734 ^b	1841 ^a	1842 ^a	1870 ^a	1847 ^a	9.28
		Feed/gain (g/g)	1.966 ^a	1.833 ^b	1.769 ^c	1.729 ^{cd}	1.713 ^d	1.685 ^d	0.006
		Breast meat (%)	15.73 ^b	17.50 ^a	17.26 ^a	17.35 ^a	17.39 ^a	17.54 ^a	0.26
Lys	ISA	Weight gain (g)	1385 ^c	1503 ^b	1543 ^{ab}	1540 ^{ab}	1557 ^a	1549 ^{ab}	7.58
		Feed/gain (g/g)	1.982 ^a	1.846 ^b	1.755 ^d	1.796 ^c	1.719 ^e	1.702 ^e	0.005
		Breast meat (%)	14.87 ^c	15.95 ^b	16.91 ^a	17.13 ^a	16.81 ^a	17.03 ^a	0.24
Lys	Ross	Weight gain (g)	1364 ^c	1554 ^b	1652 ^a	1682 ^a	1666 ^a	1694 ^a	4.62
		Feed/gain (g/g)	1.928 ^a	1.832 ^b	1.763 ^c	1.722 ^c	1.724 ^c	1.714 ^c	0.005
		Breast meat (%)	14.09 ^c	15.75 ^b	16.72 ^a	17.03 ^a	16.81 ^a	16.42 ^a	0.24
Thr	ISA	Weight gain (g)	1396 ^b	1558 ^a	1563 ^a	1572 ^a	1567 ^a	1542 ^a	32.60
		Feed/gain (g/g)	1.939 ^a	1.764 ^b	1.730 ^b	1.731 ^b	1.724 ^b	1.730 ^b	0.008
		Breast meat (%)	16.05 ^b	16.98 ^a	17.05 ^a	16.99 ^a	16.98 ^a	17.02 ^a	0.21
Thr	Ross	Weight gain (g)	1447 ^b	1770 ^a	1795 ^a	1826 ^a	1834 ^a	1793 ^a	8.86
		Feed/gain (g/g)	1.894 ^a	1.726 ^b	1.682 ^c	1.671 ^c	1.669 ^c	1.670 ^c	0.004
		Breast meat (%)	15.97 ^b	17.47 ^a	17.95 ^a	17.79 ^a	17.88 ^a	17.92 ^a	0.075
Trp	Ross	Weight gain (g)	1839 ^a	1836 ^a	1852 ^a	1859 ^a	1856 ^a	1847 ^a	6.30
		Feed/gain (g/g)	1.709 ^a	1.731 ^a	1.732 ^a	1.707 ^a	1.707 ^a	1.735 ^a	0.003
		Breast meat (%)	17.48 ^a	17.48 ^a	17.42 ^a	17.58 ^a	17.56 ^a	17.58 ^a	0.18
Arg	Ross	Weight gain (g)	1536 ^b	1642 ^a	1640 ^a	1674 ^a	1698 ^a	1706 ^a	6.25
		Feed/gain (g/g)	1.855 ^a	1.783 ^b	1.752 ^{bc}	1.725 ^c	1.691 ^d	1.687 ^d	0.004
		Breast meat (g)	344.9 ^c	367.2 ^b	357.4 ^{bc}	379.9 ^{ab}	378.8 ^{ab}	400.4 ^a	8.68
Ile	ISA	Weight gain (g)	1683 ^a	1721 ^a	1706 ^a	1729 ^a	1724 ^a	1731 ^a	5.92
		Feed/gain (g/g)	1.849 ^a	1.770 ^b	1.766 ^b	1.778 ^b	1.770 ^b	1.762 ^b	0.004
		Breast meat (%)	14.92 ^b	15.58 ^a	15.69 ^a	15.63 ^a	15.78 ^a	15.65 ^a	0.20
Val	ISA	Weight gain (g)	1345 ^c	1476 ^b	1551 ^a	1557 ^a	1543 ^a	1563 ^a	33.50
		Feed/gain (g/g)	1.937 ^a	1.770 ^b	1.754 ^b	1.720 ^b	1.719 ^b	1.710 ^b	0.009
		Breast meat (%)	16.78 ^a	16.98 ^a	17.21 ^a	17.27 ^a	17.01 ^a	17.09 ^a	0.23

a-d means within rows with different superscripts are significantly different ($P < 0.05$)

Table 4. Lysine requirement for optimum performance estimated by exponential regression analysis at 95% of the asymptotic response

Lys	Estimated requirement (g/kg)					
	Ross			ISA		
	Weight gain	Feed/gain	Breast meat (%)	Weight gain	Feed/gain	Breast meat (%)
Total	10.9	12.3	10.3	10.1	12.2	11.0
TFD	10.2	11.6	9.6	9.4	11.5	10.3

IV. CONCLUSIONS

The applied regression model has a significant impact on the calculated ideal AA profile. The broken line model is appropriate for determining the ideal AA profile but the exponential model is preferable for fixing optimum dietary lysine specifications. Lys specifications have to be assessed carefully when applying ideal AA profile in feed formulation matrix.

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EFFECT OF PROTEIN AND LINOLEIC ACID ON EGG WEIGHT OF THREE LAYER STRAINS

D. ROBINSON and M.J. DATUGAN

Egg weight is known to respond to changes in the concentrations of protein and linoleic acid in the diet of the laying hen. At practical levels of inclusion, protein tends to affect egg weight and egg number almost equally (Morris and Gous, 1988). Increasing the dietary linoleic acid level increases egg weight but not egg number in some strains of layer (Mannion *et al.*, 1992). Imported strains tend to lay eggs which are too large for current consumer requirements and may therefore benefit from nutritional regimens which aim to reduce average egg weight.

Two dietary levels of linoleic acid each at two levels of lysine (and protein) were compared for their effects on performance of three strains of laying hens. The nominal dietary concentrations of lysine and linoleic acid in the approximately isocaloric (11.6 MJ/kg) diets are shown in the table. The three strains of layer were Baiada Isabrown, Hy-line CB and Hy-line Tint. Each of the twelve combinations of strain and dietary treatment was represented by twelve groups of eight birds (four adjacent two-bird cages) distributed in randomised blocks throughout an open-sided laying shed. Performance parameters were measured from 16 to 52 weeks of age and the dietary treatment means for this period are recorded below:

Lysine (protein) g/kg	Linoleic acid g/kg	Egg wt g	Egg number	Egg mass kg	Feed intake g/bird/d	Feed/egg mass ratio kg/kg
7.1 (165)	8.5	56.76	177.1	10.07	105.8	2.667
7.1 (165)	24.0	57.63	177.2	10.22	107.1	2.654
8.2 (183)	8.5	57.53	179.4	10.33	106.6	2.615
8.2 (183)	24.0	58.29	176.9	10.31	105.6	2.601

There were large differences ($P < 0.001$) between strains in respect of egg number, egg weight, egg mass, feed intake and feed efficiency. Although there were no other significant main effects, egg weight tended to be higher at the higher levels of lysine and linoleic acid. The apparent responses in egg weight to the two nutrients were additive in the Isabrown and CB strains but not in the Tint strain, for which increases in both the lysine and the linoleic acid levels produced no further improvement in egg weight compared with increasing the level of either nutrient alone. In the Isabrown strain egg weight was higher ($P < 0.05$) but hen-day egg number was depressed by seven eggs ($P < 0.05$) on the higher linoleic acid diet, resulting in a lower total egg mass. The Isabrowns also showed higher ($P < 0.05$) egg weight and lower ($P < 0.05$) feed intake and feed/egg mass ratio when given diets with the higher lysine level.

These results suggest that changes to dietary lysine and linoleic acid levels within a practical range have small, usually independent effects on egg weight, but that increases in egg weight due to linoleic acid may sometimes be at the expense of egg number.

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Queensland Poultry Research & Development Centre, P.O. Box 372, Cleveland, Qld 4163.

USE OF DRIED DUCKWEED IN DIETS FOR BROWN EGG LAYERS

J.V. NOLAN, R. BELL, W. BALL and E. THOMSON

Aquatic plants of the duckweed family (*Lemnaceae*) provide a means of removing unwanted minerals from sewage and factory effluent. They grow rapidly and under ideal conditions may produce, annually, more than 30 tonnes of digestible, protein-rich dry matter (DM) per hectare. This protein has an amino acid profile that is similar to that of soyabean meal. Thus, the water clean-up process may also provide nutritious animal feed.

The value of duckweeds as sources of protein, minerals and yolk pigments in diets for layers has been demonstrated in relatively short-term trials (Haustein *et al.* 1990; Nolan *et al.* 1997). In this study, egg production of newer brown egg layers was recorded over a period of two months when they were given diets with or without duckweed.

Two strains of layers (Tegel Brown Egg layers and Tegel Hi-Sex, a total of 108 hens 55 weeks of age) were randomly allocated to single-bird cages (banks of 6) distributed uniformly in an open shed. All hens were given commercial crumbles for 4 weeks before being allocated to their dietary treatments, *viz.* Diet 1 Ridley's AgriProducts commercial layer crumble (Control 1); Diet 2, layer crumble (Control 2); Diet 3, Duckweed layer crumble with 10% duckweed (DW). The three diets were formulated to have the same specifications, *i.e.* 10.8 MJ/kg DM, 16.7 % crude protein, 3.8 % Ca, 0.59% available P).

Feed intake and egg production were recorded for 8 weeks. Egg weights, and shell and internal characteristics were determined, within 24 h, on eggs collected on two successive days at the end of the trial. Results are given in the Table.

	Strains		Diets			Significance	
	Hi-Sex	Brown n	Ridley's diet	Control diet	DW diet	Strain	Diet
Intake (gDM/d)	142	140	137	139	147	NS	NS
Hen-day prod.(%)	88.4	86.0	90.7	84.9	85.9	NS	NS
Egg weight (g)	67.4 ^a	62.4 ^b	63.6 ^a	64.8 ^a	66.2 ^b	<0.001	<0.01
Shell thick (µm)	382 ^a	369 ^b	379	369	378	<0.05	NS
Haugh units	74.0	73.4	72.6	73.9	74.6	NS	NS
Yolk colour Roche	12.6	12.7	12.2 ^a	12.5 ^a	13.2 ^b	NS	<0.001

^{a,b} Means with different superscripts differ significantly (two-way analysis of variance).

There were some significant effects of diet (and strain), but no significant interactions. Feed intake and production was excellent on all diets and adverse clinical effects of the duckweed were not evident. Birds on the duckweed-containing diet produced slightly heavier eggs with attractive but darker yolks. We conclude that duckweed is a useful protein, mineral and pigment source in diets for layers. It appears to contain no toxic or anti-nutritional factors: it would, however, be pertinent to further evaluate duckweed-containing diets for layers over a 50-week laying period.

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Division of Animal Science, University of New England, Armidale NSW 2351.

EFFECTS OF DIFFERENT CEREAL GRAINS ON EGG AND EGG SHELL QUALITY IN LAYING HENS

J.R. ROBERTS, W. BALL and A. LEARY

This study was part of a project investigating the factors contributing to wet droppings in Isa Brown laying hens. Layer rations were formulated based on a range of cereal grains: sorghum, triticale, barley or wheat and compared with a standard commercial layer ration. Birds were housed, two to a cage, in a commercial laying house at the University of New England "Laureldale" Farm. Rations were fed to the birds for 10 weeks prior to the collection of sixty eggs from each of the five diets for detailed analysis (a total of 300 eggs). The following measurements were made on the eggs: egg weight, gross egg shell defects, egg shell pigmentation (by reflectivity), egg length and breadth, eggshell breaking strength (by quasi-static compression), shell weight and shell thickness (using a dial comparator gauge). Shape index (breadth x 100/length) and percentage shell (shell weight : egg weight) were calculated. Internal egg quality was assessed by measuring the Haugh units of the albumen and the Roche score of the yolk.

Egg weight was highest for the wheat diet and lowest for the triticale with the other diets producing intermediate sized eggs. The sorghum diet produced egg shells with the greatest breaking strength whereas the weakest shells occurred on the barley diet. Shell weight and shell thickness were greatest on the control and sorghum diets and lowest on the triticale and barley diets. A similar trend occurred with the percentage shell (shell weight : egg weight ratio). Yolk colour was greatest for the control and sorghum diets and lowest for the barley diet. Shell reflectivity (colour) and Haugh Units were not significantly different between diets.

These results indicate that the type of cereal grain on which a layer ration is based can affect egg shell quality. In addition, the amount of yolk pigment added to the ration needs to be adjusted in accordance with the type of cereal grain in order to optimise yolk colour.

Table 1. Egg and egg shell quality for diets based on different cereal grains.

	Control	Sorghum	Triticale	Barley	Wheat
Egg Wt g	^{ab} 63.5±0.5	^b 62.9±0.6	^c 60.9±0.5	^{ab} 63.4±0.6	^a 64.9±0.5
Shell Reflectivity %	35.6±0.6	36.5±0.8	37.0±0.7	36.4±0.5	36.2±0.8
Breaking Strength N	^{ab} 31.0±1.3	^a 32.5±1.3	^{bc} 28.3±1.1	^c 26.8±1.1	^{abc} 29.2±1.1
Shell Wt g	^a 5.97±.06	^a 5.96±.08	^c 5.34±.07	^b 5.60±.06	^a 5.85±.06
Shell Thickness µm	^a 384.7±3.4	^a 386.6±4.2	^c 356.9±3.6	^c 361.9±3.4	^b 373.9±3.2
% Shell	^a 9.42±.11	^a 9.49±.10	^b 8.78±.10	^b 8.86±.09	^b 9.01±.08
Yolk Colour	^a 11.6±.08	^a 11.7±.09	^b 10.6±.15	^c 9.9±.13	^b 10.7±.11
Haugh Units	79.1±1.5	80.1±1.3	77.7±1.6	77.0±1.4	77.5±1.4

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Division of Animal Physiology, School of Rural Science and Natural Resources, Faculty of the Sciences, University of New England, Armidale, NSW 2351.

ULTRASTRUCTURAL ANALYSIS OF EGG SHELL LAMINAE

J. RUIZ¹, P. GROVES², P. GLATZ³, J. MELDRUM⁴ and C. LUNAM¹

The ultrastructural integrity of the eggshell is crucial to embryonic development. In addition to providing physical protection, gaseous exchange across the eggshell is essential for successful hatching and for optimal growth of the chick. Gaseous porosity is dependent on the total cross-sectional area of the shell and on the number of pores within the calcified region (Ar *et al.*, 1974). Although it is known that the eggshell is comprised of four discrete calcified laminae, limited information is available concerning the proportion of the individual lamina within the eggshell across different broiler strains. The overall aim of this work is to compare the relative proportion of the individual calcified layers in two imported lines of meat birds having different hatchability levels. This initial work was undertaken to determine whether the cross-sectional lengths of the calcified laminae within the eggshell are conserved within a strain by comparing the proportion of each calcified lamina between grandparent and parent levels of a single commercial broiler breeder line.

Sixteen eggs were randomly selected from a single collection day for each of the grandparent and parent lines. For each egg, shell thickness was measured from three equatorial pieces (without membranes) using a thickness gauge. To facilitate ultrastructural viewing, all pieces of shell were pre-treated with liquid nitrogen and then freeze-snapped. The cross-sectional length of each lamina was measured using a Siemens ETEC scanning electron microscope. The relationship between the cross-sectional length of each of the four laminae to the total cross-sectional length of the calcified region was determined using Pearson's correlation test. Differences in the proportion of each of the four laminae between the grandparent and parent lines was assessed using analysis of variance (SPSS Inc., 1995). No differences were observed in the proportions of any of the four calcified laminae between grandparent and parent levels; *p* values are given in the table below. The mammillary and palisade layers comprised at least 85% of the total calcified region.

Laminae	% Total Cross-Sectional Length		
	Parent line	Grandparent line	<i>P Value</i>
Mammillary	26.8 - 31.1 (<i>r</i> : 0.62)	25.3 - 31.7 (<i>r</i> : 0.73)	0.83
Palisade	63.8 - 70.0 (<i>r</i> : 0.95)	62.1 - 70.2 (<i>r</i> : 0.98)	0.69
Vertical Crystal	3.1 - 6.9	2.3 - 6.2	0.70
Cuticle	0.7 - 3.1	0.7 - 2.0	0.10

r: Pearson Correlation Coefficient, significant at *P*<0.01.

Lower hatchability in eggs from hens at an early stage of lay tends to be associated with an increase in shell thickness compared with that at a later stage of lay. Further studies in which the proportion of the mammillary and palisade laminae of eggs from hens at different ages will determine if the reduction in shell thickness is accompanied by a change in the relative proportion of the calcified laminae.

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¹ Department of Anatomy and Histology, Flinders University, Bedford Park, SA 5042.

² Baiada Poultry Pty Ltd, PO Box 21, Pendle Hill, NSW 2145.

³ SARDI, Roseworthy Campus, University of Adelaide, Roseworthy, SA 5371.

⁴ HiChick Breeding Company, PO Box 821, Gawler, SA 5118.

THE INFLUENCE OF XYLANASES ON ENERGY AND AMINO ACID AVAILABILITY IN WHEAT USED IN BROILER DIETS

W.D. COWAN¹ and D.R. PETTERSSON²

Mono component xylanases are now available and they possess improved technological characteristics and possibly also zoo-technological properties compared to polyvalent enzymes. This study was made to examine the influence of a mono component xylanase (from *Thermomyces lanuginosa*) on energy and amino acid availability in wheat and to compare it with a reference xylanase, (from *Humicola insolens*). The basal diet was composed of sorghum (560 g/kg), soya (320 g/kg) and animal fat (60 g/kg).

For determination of the nitrogen-corrected, apparent metabolisable energy (AMEn) of wheat, 500 g/kg basal diet was combined with 500 g/kg wheat. Amino acid digestibility was determined quantitatively. The influence of xylanases upon nutrient availability was determined by supplementing the diets with different levels (0-400 FXU/kg of diet) of the mono component xylanase or with 400 FXU/kg of diet of the reference xylanase.

Supplementation of the wheat with 400 FXU/kg of the reference xylanase increased the wheat AMEn to a level equivalent to that obtained with 200 FXU/kg of the mono component xylanase. Addition of 200 FXU/kg of the mono component xylanase resulted in an amino acid digestibility which was higher than that obtained with 400 FXU/kg of the reference xylanase. Thus, a mono component enzyme has been shown to be more effective at increasing nutrient availability than the reference xylanase.

Table 1. Influence of Xylanase on wheat AMEn content.

Xylanase	Dose /kg diet	AMEn (MJ/kg)
Control	0 FXU	11.81
Mono component	100 FXU	12.24
Mono component	200 FXU	12.51
Mono component	400 FXU	12.80
Reference	400 FXU	12.52

Table 2. Influence of Xylanase on amino acid digestibility (%) in wheat.

Amino Acid	Control	Reference	Mono
Lysine	67	70	76
Methionine	72	79	80
Tryptophan	67	73	74
Threonine	63	71	75
iso-Leucine	74	79	79
Relative average digestibility	100a	108.5b	113.5b

Results are expressed on an "as is" basis. Means with different subscripts are significantly different at $P < 0.005$.

¹ Novo Nordisk S.A. 282 Chartridge Lane, Chesham, Bucks HP5 2SG, UK.

² Novo Nordisk A/S, DK 2880 Bagsvaerd, Denmark.

ENZYMES FOR MAIZE-BASED LAYER DIETS

D. CRESWELL¹, M. PACK² and D. ROBINSON³

Improved performance and economics from wheat and barley based layer diets with xylanase and beta-glucanase enzymes have been reported by Kumar *et al.* (1997). The present study tested an enzyme product with xylanase, amylase, and protease activities (Avizyme 1500™, Finnfeeds International Ltd) in layer diets based on maize. The trial was conducted at Queensland Poultry Research & Development Centre using 200 74-week-old ISABrown layers. Birds were housed in single cages, allowing 50 replicates for each of the four treatments. Diets were based on maize (571 and 517 g/kg), soyabean meal, wheat pollard and tallow. A high specification (HS) control diet was formulated to contain 11.5 MJ/kg AME, 17.0g/kg protein, 0.94g/kg lysine and 0.65g/kg met+cys. A medium specification (MS) diet was formulated with 5% lower AME, protein and amino acid minimums. There was 50 and 150 g/kg wheat pollard in the HS and MS diets respectively. These two diets were fed without or with the enzyme (0.75 kg/t.) The trial lasted 12 weeks and results are shown below. Egg production per hen housed was not statistically analyzed.

Treatments	HS diet		MS diet		Enzyme effect (P<)
	E -	E +	E -	E +	
Egg prod. per hen day, %	77.6 ^a	78.7 ^a	70.7 ^b	75.6 ^a	0.04
Egg prod. per hen housed, %	75.3	78.0	69.4	74.3	-
Egg weight, g	67.2 ^a	67.0 ^a	64.6 ^b	67.1 ^a	0.03
Egg mass, g/d	52.1 ^a	52.7 ^a	45.7 ^b	50.7 ^a	0.02
Feed intake, g/d	114.7 ^b	113.0 ^b	115.0 ^{ab}	117.3 ^a	0.55
FCR, g feed/g egg	2.20 ^a	2.14 ^a	2.52 ^b	2.31 ^a	0.05
Mortality, %	4	2	4	2	-

^{ab} Values in the same row with a common superscript are not different (P<0.05)

Enzyme supplementation improved egg production, egg weight, daily egg mass, and feed conversion of birds on the MS diet (11% increase in daily egg mass). It is likely that these effects of enzyme supplementation on production are achieved through an improvement in the energy utilization of the maize and wheat pollard.

These results indicate it is possible to improve performance from maize based layer diets with the use of exogenous enzymes and that by modifying dietary specifications the benefits from added enzymes can be maximized.

Kumar, A., Dingle, J. and Creswell, D. (1997) In: *Recent Advances in Animal Nutrition in Australia* p 226. Eds. J.L. Corbett, M. Choct, J.V. Nolan, J.B. Rowe. University of New England, Armidale.

¹ Creswell Livestock Consultants, Mosman, NSW 2088.

² Finnfeeds International Ltd, Marlborough, United Kingdom.

³ Queensland Poultry Research and Development Centre, Alexandra Hills, Qld 4161.

INFLUENCE OF MICROBIAL PHYTASE ON APPARENT METABOLISABLE ENERGY AND AMINO ACID DIGESTIBILITY IN BROILERS

S. CABAUG¹, V.RAVINDRAN¹, W.L. BRYDEN¹ and P.H. SELLE²

The use of microbial phytase to release phytate-bound phosphorus (P) in plant feed ingredients has attracted considerable attention in recent years. The effectiveness of microbial phytase in improving overall P availability for growth and bone mineralisation in broilers is now well documented. However, published data on the influence of added phytase on energy utilisation and amino acid digestibility are limited. The influence of microbial phytase (Natuphos®, BASF AG, Germany) on apparent metabolisable energy (AME) and apparent ileal amino acid digestibility (AIAD) are reported in this paper.

Nine hundred 7-day old male broiler chicks were used in a 3 x 3 x 2 factorial arrangement of treatments with five replicates (10 chicks/pen) to study the response to three levels of phytase (0, 400 and 800 FTU/kg diet) when given in combination with three levels of phytic acid (PA; 10.4, 13.2 and 15.7 g/kg) and two levels of non-phytate P (nP; 2.3 and 4.5 g/kg). Details of the study have been previously reported (Cabaug *et al.*, 1997). The diets were fed from day 7 to 25. The classical total collection method was employed during the last 3 days to determine the AME values of the diets. On day 25, digesta contents from the terminal ileum were collected and processed. Samples of diets and digesta were analysed for acid-insoluble ash and amino acids, and the AIAD values were calculated.

Increasing levels of PA had no influence, but main effects ($P < 0.001$) of nP and phytase were observed for AME values. An nP x phytase interaction ($P < 0.001$) was also observed for AME, indicating that the effects of phytase on AME was dependent on dietary nP levels. The AME of diets containing low levels of nP were largely unaffected by added phytase, whereas consistent improvements of 5-6% were seen in diets with adequate levels of nP. The AME of low and adequate nP diets with 0, 400 or 800 FTU phytase/kg were 13.36, 13.58 and 13.49, and 12.66, 13.31 and 13.45 MJ/kg dry matter, respectively. Main effects of PA ($P < 0.03$ to 0.001) and phytase ($P < 0.01$) and an interaction of nP x phytase ($P < 0.01$ to 0.08) were observed for the digestibility of most essential amino acids. Addition of phytase improved amino acid digestibilities at both nP levels, but the responses were greater in diets containing low level of nP. The mean percentage increases in digestibility in diets containing low nP diets plus phytase were as follows: lysine, 3.0; arginine, 3.7; threonine, 6.9; isoleucine, 4.6; leucine, 6.4; valine, 5.4; phenylalanine, 5.1; and histidine, 4.6. The corresponding increases in diets containing adequate nP diets plus phytase were 1.4, 1.8, 3.0, 2.4, 2.9, 2.4, 2.7 and 1.8, respectively. These findings emphasise the relevance of phytate(P)-protein complexes and the anti-nutritional effects of PA on amino acid availability in practical broiler diets. The mechanism underlying the energy effect of added phytase in adequate nP diets is unclear, but it appears to be independent of the protein effect of the enzyme. The present results demonstrate the beneficial effects of microbial phytase on energy metabolism and protein digestion in chickens.

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¹ Department of Animal Science, The University of Sydney, Camden, NSW 2570.

² BASF Animal Nutrition, Auburn, NSW 2144.

A SIMPLE METHOD FOR THE MEASUREMENT OF ENDOGENOUS
β-GLUCANASE ACTIVITY IN WHEAT

D.L.E. WATERS and M. CHOCT

The nutritive value of cereal grains for poultry generally changes during storage. It is believed that this change is due to the activity of endogenous glycanases which degrade the viscous non-starch polysaccharides. However, these enzymes are present at such low levels in ungerminated wheat that measurement of their activity is difficult (Lee and Ronalds, 1972). We report here a simple procedure for the measurement of endogenous β-glucanase activity. Ten wheats were milled in a Cyclotec 1093 Sample Mill with a 0.5 mm screen. Following this, 4 g of flour from each sample was extracted in 100 ml of buffer at 40°C for 20 minutes with occasional mixing. A 24 ml subsample of each extract was centrifuged at 2000 g for 10 minutes and then 300 μl of the supernatant was added to each of three 3 ml replicates of a buffered 1% barley β-glucan (Megazyme, Australia) solution in 0.02% sodium azide and incubated at 40°C with shaking. Aliquots were taken at 0 hours, 24 hours and 48 hours and placed on ice. The viscosity of these solutions was immediately determined using a Brookfield Model DV-III Rheometer at a constant temperature of 25°C. The control was 3 ml of buffered substrate with 300 μl of extraction buffer added. The results were as follows:

Sample	0 Hours		24 hours		48 Hours	
	Viscosity mPa.s	% Change	Viscosity mPa.s	% Change	Viscosity mPa.s	% Change
Wheat 1	6.67	0	4.43 ^e	-34 ^f	3.63 ^f	-46 ^f
Wheat 2	6.82	0	5.79 ^c	-15 ^c	5.15 ^c	-24 ^c
Wheat 3	6.61	0	4.75 ^d	-28 ^d	4.03 ^d	-39 ^d
Wheat 4	6.60	0	4.52 ^e	-32 ^e	3.79 ^{ef}	-43 ^e
Wheat 5	6.80	0	5.83 ^{bc}	-14 ^c	5.29 ^{bc}	-22 ^c
Wheat 6	6.85	0	5.83 ^{bc}	-14 ^c	5.10 ^c	-25 ^c
Wheat 7	6.74	0	4.76 ^d	-29 ^d	4.00 ^{de}	-40 ^d
Wheat 8	6.78	0	5.66 ^c	-16 ^c	5.00 ^c	-26 ^c
Wheat 9	6.74	0	5.67 ^c	-16 ^c	5.09 ^c	-24 ^c
Wheat 10	6.82	0	5.98 ^b	-12 ^b	5.47 ^b	-20 ^b
Control	6.80	0	6.76 ^a	-1 ^a	6.84 ^a	+1 ^a

a-f unlike superscripts within a column are significantly different (P<0.05).

Viscosity reduction between different wheats varied widely suggesting that the endogenous β-glucanase activity also differs. The practical implication of this finding for the poultry industry in terms of the use of cereal grains as energy sources will be examined in future work.

Lee, J.W. and Ronalds, J.A. (1972). *J. Sci. Fd. Agric.* **23**: 199-205.

Division of Animal Science, University of New England, Armidale, NSW 2351.

LAYER PRODUCTION INCREASED BY ENZYME SUPPLEMENTATION OF SORGHUM BASED DIETS

A. KUMAR¹, J.G. DINGLE¹ and D. CRESWELL²

In recent years scientists have shown that enzyme supplementation of barley or wheat based diets has improved layer production and net returns (Kumar and Dingle, 1995 and Kumar et al., 1997). The success of enzyme supplementation of wheat and barley based broiler and layer diets stimulated interest in using enzymes in sorghum based layer diets. The present study was undertaken to find the effect of a multi-enzyme product (Avizyme 1500) containing xylanase, protease and amylase on laying hen production when added to diets based on sorghum grain.

A total of 216 HiSex Brown layers aged 32 weeks were assigned to cages at two birds per cage, with 108 birds for each of the two dietary treatments. There were 54 replicates/diet for egg production and 9 replicates/diet for feed intake measurement. The diet consisted of sorghum (65.8%), barley (5%), meat and bone meal (6.9%), cotton seed meal (6.9%), canola meal (6%) and vitamins, minerals and amino acids (9.4%) to provide 11.5 MJ of ME and 161g protein per kg of feed. Feed was offered *ad libitum* in mash form with or without the multi-enzyme product, Avizyme 1500, at the rate of 0.75 kg/t of feed. Egg production and weight, food consumption and mortality were recorded for 17 weeks. A summary of the results is shown in the Table.

	Control	+Avizyme 1500 (0.75 g/kg)	SEM
Egg prod.(HH) %	76.2 ^b	82.1 ^a	1.81
Feed intake (g/d)	114.8	118.2	1.82
FCR	2.40 ^a	2.27 ^b	0.03
Egg weight (g)	61.9	62.5	0.41
Egg mass (g/d)	47.2 ^b	51.5 ^a	1.18
Mortality (%)	3.7	2.8	1.98
Net profit ¹	0.58 ^b	0.63 ^a	0.01

Means within rows with different superscripts are significantly different (P<0.05).

¹ Egg income minus feed cost/hen/week (AUD) (cost of enzyme included at AUD 4.125/t feed).

Whilst egg production was generally lower than expected, enzyme supplementation of the diet significantly improved egg production by 7.7 %, daily egg mass by 9% and feed efficiency by 5.4% during the 17 weeks trial. These improvements were probably due to an increase in nutrients released from sorghum and protein meals by the action of the enzymes in Avizyme 1500 on arabinoxylans, starch and proteins. Feed intake and mortality were not significantly affected by enzyme addition. Enzyme supplementation increased the mean net profit/bird by AUD 0.85 during the 17 weeks of the trial (P<0.05).

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¹ Department of Animal Production, The University of Queensland, Gatton, 4345.

² Creswell Livestock Consultants, Mosman, NSW 2088.

NEUROMA FORMATION IN LAYERS AFTER RE-TRIMMING

C.A. LUNAM¹, P.C. GLATZ² and J.L. BARNETT³

Neuromas are bundles of disarrayed nerves that form as part of the normal regenerative process following injury. Neuromas pose significant welfare implications as they can spontaneously discharge action potentials resulting in chronic pain. In addition, nerve fibres associated with neuromas have a decreased threshold to noxious stimuli and undergo allodynia, a condition in which normally non-noxious stimuli become painful. Previous work has demonstrated that beak trimming at hatch results in the formation of neuromas, regardless of the amount of tissue removed. However, neuromas persist to adulthood only in hens after severe trimming (Lunam *et al.*, 1996). This work is part of a study of the time course of neuromal development and resolution after moderate beak trimming at hatch. The effect of re-trimming on the formation and resolution of neuromas is also being investigated.

At hatch, layers were allocated into three groups: group 1, removal of half the upper beak and one third of the lower beak at hatch, re-trimmed (2mm removed from each of the upper and lower beak) at 14 weeks; group 2, trimmed at hatch only and group 3, non beak-trimmed controls. Four layers were randomly selected from each of the three groups and euthanised by cervical dislocation at 28 weeks of age. The upper and lower beaks were processed for histopathology. Sagittal sections, collected at 200 micrometer intervals through each beak, were stained either with silver for visualisation of the presence or absence of neuromas, or with haematoxylin and eosin for general tissue structure.

Neuromas were observed in all trimmed and re-trimmed beaks. These were marginally more extensive after re-trimming. Neuromas consisted of swirling masses of nerve fibres as well as small foci of micro-neuromas encapsulated in a connective tissue sheath. Neuromas were more extensive in the upper beaks compared to those in the lower beaks. Individual nerve fibres often reached the dermal-epidermal margin and penetrated into the tip of the beak stump. In 3 of the 4 upper beaks that had been trimmed only at hatch (group 2) the neuromal masses were confined to the ventral dermis adjacent to the premaxillary bone. In these beaks, nerve bundles within the dermis between the neuroma and epidermis appeared similar to those in the non-trimmed control beaks. Although fewer than in non-trimmed control beaks, sensory receptors were present in all single trimmed and re-trimmed beaks. Receptors were often present in the distal dermis near the tips of all trimmed and re-trimmed lower beaks.

These results demonstrate that neuromas are present 28 weeks after moderate trimming at hatch. The extent and distribution of neuromas however, compared to that in beaks 10 weeks after moderate trimming at hatch (Lunam *et al.*, 1996), suggest much of the neuromal mass has been resorbed by 28 weeks. The marginal increase in the extent of neuromas 14 weeks after retrimming, as well as the presence of sensory receptors close to the beak tip in re-trimmed beaks, suggests that re-trimming of 2mm causes minimal perturbation of nerves and general tissue structure.

Lunam, C.A., Glatz, P.C. and Hsu, Y-J. (1996). *Aust. Vet. J.* **74**: 46-49.

¹ Department of Anatomy & Histology, Flinders University, Bedford Park, SA 5042.

² South Australian Research and Development Institute, Roseworthy, SA 5371.

³ Animal Welfare Centre, Victorian Institute of Animal Science, Werribee, Vic. 3030.

PAIN THRESHOLDS IN BEAK TRIMMED BIRDS

E.C. JONGMAN¹, J.L. BARNETT¹, P.C. GLATZ² and C.A. LUNAM³

This experiment assessed long term pain in beak trimmed poultry by measuring behavioural responses in birds to stimuli at the time when neuromas are in the process of resolving, 10 weeks after trimming at hatch. The birds were tested at 10 weeks of age and again at 20 and 60 weeks. This paper describes the first testing period when birds were 10 weeks old.

Of a total of 60 birds, 40 were moderately beak trimmed at hatch and 20 birds were non trimmed controls. The birds were housed in individual cages. Individual birds were then placed into a test cage and the following tests to measure changes in pain perception as a result of beak trimming were conducted. 1) *Drinking water at 20°C*: after deprivation of drinking water for 15-16 h prior to testing, individual birds were placed in the test cage, and drinking behaviour (pecking at water, drinking and head shakes) during 5 min was recorded on video. 2) *Drinking water at 45°C*: similar to test 1, but with water at 45°C. 3) *Pecking at food*: after food deprivation for 1-1.5 h prior to testing, individual birds were placed in the test cage and the force applied when pecking a thin layer of food was measured (Dynamometer UF1, range -100 to +100 g). During the test, birds were recorded on video to ensure the peak force recorded coincided with a pecking event. 4) *Pecking at a red disk*: as in test 3 with pecking behaviour and peak pecking force at a red disk being recorded.

Results are shown below. Trimmed birds showed more head shakes in test 2 (water at 45°C) and applied a lower peak force when pecking at a red disk.

Behaviour observed	Intact birds	Birds trimmed at hatch
Pecking at water (20°C)	5.6 ^x	11.9 ^y
Drinking water (20°C)	5.1 ^x	11.1 ^y
Head shakes (20°C)	0.18	0.40
Drinks per peck (20°C)	0.25 ^a	0.58 ^b
Head shakes per peck (20°C)	0.024	0.028
Pecking at water (45°C)	5.1	7.7
Drinking water (45°C)	3.0	3.3
Head shakes (45°C)	0.24 ^c	1.57 ^d
Drinks per peck (45°C)	0.21	0.33
Head shakes per peck (45°C)	0.05 ^c	0.18 ^d
Pecking at food (force g)	5.1	4.0
Pecking at red disc (force in g)	2.11 ^x	1.26 ^y

^{a,b} P<0.01, ^{c,d} P<0.05, ^{x,y} P<0.1

At ten weeks of age the beak of birds trimmed at hatch was more sensitive to pain, as indicated by the higher number of head shakes and head shakes per peck when attempting to drink warm water and the lower force used when pecking at the red disk. The higher frequency of pecking and drinking of the cooler water suggests that at this age beak trimmed birds may be less effective at drinking water.

¹ Animal Welfare Centre, Victorian Institute of Animal Science, Werribee, Victoria 3030.

² South Australian Research and Development Institute, Roseworthy, SA 5371.

³ Department of Anatomy and Histology, Flinders University, Bedford Park, SA 5042.

EVALUATION OF HIGH-METHIONINE TRANSGENIC LUPINS IN BROILER DIETS

V. RAVINDRAN¹, W.L. BRYDEN¹, L.M. TABE², T.J.V. HIGGINS² and L. MOLVIG²

Lupins, in particular narrow leaved lupin (*Lupinus angustifolius*), are an important source of protein in Australian poultry diets. Their seed protein however is deficient in sulphur-containing amino acids (SAA), necessitating the addition of synthetic methionine to diets containing lupins. A transgenic lupin with higher contents of SAA has been recently developed through the introduction of a chimeric gene encoding the SAA-rich sunflower seed albumin protein to *L. angustifolius* cv Warrah (Spencer *et al.*, 1997). The nitrogen, methionine and cystine contents in the parental, non-transgenic (N-TG) and transgenic (TG) lupins were 47.5 and 47.0; 1.9 and 4.1; and 4.4 and 3.7 g/kg air dry basis, respectively. This paper describes two separate trials which evaluated the nutritive value of TG lupins for broiler chickens.

In Trial 1, N-TG and TG lupins, with hulls, were incorporated into a maize-soyabean meal diet at 250 g/kg level. All diets were balanced to contain similar levels of apparent metabolisable energy (AME), protein, lysine and SAA. The levels of synthetic methionine added to the maize-soyabean meal control, N-TG lupin and TG lupin diets were 2.2, 2.8 and 2.2 g/kg, respectively. Four replicates of ten female broiler chicks (Cobb) were fed each of the experimental diets from 6 to 20 days of age. Weight gain and feed intake were not influenced by dietary treatments, but feed/gain tended to be higher ($P < 0.09$) in birds fed lupin diets compared to those on the control diet. Feed/gain of birds fed the N-TG lupin diet were numerically higher than those fed the TG lupin diet. These results showed that the supplemental methionine required in poultry diets containing 250 g/kg lupins can be lowered by 0.6 g/kg diet (600 g/tonne) by the use of high-methionine TG lupins.

In Trial 2, the AME of lupins was determined in a classical total excreta collection assay using 24 individually-housed 5-week old male broilers (Cobb). A basal diet, containing maize and casein as the main ingredients, served as the control and the test diets were formulated by substituting (w/w) 250 g/kg of the basal diet with N-TG or TG lupins. Assuming an additive model, the AME (mean \pm SE) of N-TG and TG lupins were calculated to be 9.42 ± 0.55 and 10.18 ± 0.27 MJ/kg dry matter, respectively.

The higher AME values in TG lupins may be related to the lower contents of total non-starch polysaccharides (369 vs 417 g/kg dry matter; K.E.B. Knudsen and L. Tabe, unpublished data).

Diet type	Supplemental meth (g/kg diet)	Weight gain (g)	Feed intake (g)	Feed/gain (g/g)
Maize-soyabean meal	2.2	454	743	1.64
250 g/kg N-TG lupin	2.8	442	803	1.82
250 g/kg TG lupin	2.2	441	767	1.74
Pooled SEM		12.6	16.9	0.046

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¹ Department of Animal Science, The University of Sydney, Camden, NSW 2570.

² C.S.I.R.O., Division of Plant Industry, GPO Box 1600, Canberra, ACT 2601.

BROILER FEED FORMULATIONS WITH CANOLA MEAL BASED ON TOTAL OR DIGESTIBLE AMINO ACIDS

V. RAVINDRAN, L.I. HEW and W.L. BRYDEN

Canola meal (CM) is being increasingly used by the local feed industry, but past attempts to substitute CM for part of soyabean meal in broiler diets have resulted in less-than-expected performance because the substitutions have often failed to take into account the lower amino acid (AA) digestibility in CM. Preliminary studies from our laboratory have shown that diets formulated with 100 g CM/kg on the basis of true ileal or apparent ileal digestible lysine yielded similar bird performance. In the present study, broiler diets were formulated with graded levels of CM on a total AA vs apparent ileal digestible AA basis and evaluated in a broiler growth assay. A wheat-sorghum-soyabean meal diet, formulated to contain 12.8 MJ apparent metabolisable energy/kg, 223 g crude protein/kg, 12.5 g total lysine/kg and 0.64 g total methionine/kg, served as the control diet. Diets 2 to 4 were formulated to contain 66.6, 133.3 and 200 g CM/kg, and similar levels of total AA to those in the control diet. Diets 5 to 7 contained the same graded levels of CM, but were balanced to contain similar levels of apparent ileal digestible AA. Apparent ileal AA digestibility of all ingredients was determined prior to diet formulation. Apparent ileal digestible lysine requirement was assumed to be 87% of the total requirement, and the ideal protein concept (Baker and Han, 1994) was applied to estimate the requirements for the other essential AA. Each diet was fed to four pens of six male chicks (Cobb) from day 5 to 19 post-hatching. The performance data are summarised below.

Dietary treatment	Weight gain (g/bird)	Feed intake (g/bird)	Feed/gain (g/g)
Wheat-sorghum-soyabean meal diet (control)	472	668	1.42
<i>Formulation on total amino acid basis</i>			
66.6 g canola meal/kg	469 (99) ¹	678 (101)	1.45 (102)
133.3 g canola meal/kg	458 (97)	684 (102)	1.49 (105)
200.0 g canola meal/kg	456 (96)	678 (101)	1.49 (105)
<i>Formulation on digestible amino acid basis</i>			
66.6 g canola meal /kg	471 (100)	664 (100)	1.41 (99)
133.3 g canola meal/kg	492 (104)	689 (103)	1.40 (99)
200.0 g canola meal/kg	487 (103)	699 (105)	1.44 (101)
Pooled SEM	12.1	15.8	0.03

¹ Values in parentheses refer to bird performance relative to the control.

Dietary inclusion of 133.3 and 200 g CM/kg on a total AA basis resulted in numerically lower weight gain and higher feed/gain. Birds fed diets containing 133.3 and 200g CM/kg and formulated on a digestible AA basis had numerically higher weight gain and lower feed/gain values than those fed corresponding CM diets formulated on a total AA basis. Although none of the differences was statistically significant, the performance data suggest that formulating broiler diets on a digestible AA basis is beneficial when poorly digestible ingredients are incorporated.

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Department of Animal Science, The University of Sydney, Camden, NSW 2570.

MANIPULATION OF GLUCOSE METABOLISM IN THE BROILER CHICKEN WITH DIETARY FATTY ACIDS

R.E. NEWMAN, J.A. DOWNING, J.A. DEHON and W.L. BRYDEN

Dietary (n-3) polyunsaturated fatty acids have been shown to influence glucose metabolism in rodents and in man. Luo *et al.*, (1996) demonstrated increased rates of glucose oxidation in adipocytes from rats fed dietary n-3 polyunsaturated fat, while in man n-3 fatty acids significantly increased glucose utilisation (Waldhäust *et al.*, 1989). This preliminary study was designed to establish dietary (n-3) polyunsaturated fatty acids can influence whether glucose metabolism in broilers.

Two diets were prepared containing tallow, a source of saturated fatty acids (80 g/kg), and fish oil, a source of (n-3) polyunsaturated fatty acids (80 g/kg). These two diets had similar determined AME values and were isonitrogenous. Broiler chickens (30 days old) were randomly divided into 2 groups (n=10) and were fed the diets for 5 weeks. Jugular catheterisation was performed under general anaesthesia during week 4. The broilers were allowed 7 days post surgery to recover before sampling blood every 2 h for 8 h. Plasma glucose and triglyceride concentrations were measured by enzymic analysis. To estimate the glucose clearance rate from plasma and glucose incorporation into tissues, 2-deoxy-D-³H glucose (2DG-³H) was infused into each chicken (50 μ Ci) at the end of the study. Sequential blood samples were then taken via the indwelling catheter over a period of 1 h.

The mean plasma glucose concentrations over the 8 h period were elevated, but not significantly, in the broilers fed the fish oil diet (12.83 \pm 0.59 mmol/L, vs. 12.07 \pm 0.56 mmol/L). The mean plasma triglyceride concentrations were lower in broilers fed the fish oil, but again not significantly (1.97 \pm 0.21 mmol mmol/L vs. 2.15 \pm 0.16 mmol/L). The rate of 2DG-³H clearance from the plasma of broilers fed fish oil was significantly greater (P<0.05) than for the broilers fed tallow. The 2DG-³H was cleared after 15 min in the broilers fed fish oil compared to 30 min for the broilers fed tallow. There was a greater rate of 2DG-³H uptake into the breast and thigh muscles of the broilers fed fish oil compared to broilers fed tallow (breast muscle 4100 cpm/g tissue, and 2500 cpm/g tissue respectively; thigh muscle 4700 cpm/g tissue and 2900 cpm/g tissue respectively). The uptake of 2DG-³H into the liver was greater in the broilers fed tallow compared to those fed fish oil (9100 cpm/g tissue and 7300 cpm/g tissue). There was no difference between the two dietary groups in the uptake of 2DG-³H into the abdominal fat pad.

These data suggest that the type of fat in broiler diets can influence glucose metabolism. The inclusion of fish oil in place of tallow increases the rate at which glucose is cleared from blood and also increases the rate of glucose uptake into muscle tissue.

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Department of Animal Science, The University of Sydney, Camden, NSW 2570.

IMPAIRED PERFORMANCE OF CHICKENS FED SORGHUM INFECTED WITH
ERGOT (*CLAVICEPS AFRICANA*)

P.F. MANNION¹ and B.J. BLANEY²

Sorghum ergot (*Claviceps africana*) was first identified in Australia in April 1996, and within six months had been found in all sorghum producing regions in Queensland and New South Wales. Ergot fungi infect the plant during flowering and after a growth cycle characterised by release of plant sap (honeydew) and spores, the fungus develops hard sclerotia (ergots) containing alkaloids which either fall to earth or are harvested with grain.

Sorghum ergot produces a different range of alkaloids than the ergot of rye (*C. purpurea*). Rye ergot reduces the growth of chickens at concentrations above 16 g/kg and, through impaired circulation, produces gangrene of the toes, beak and comb (Bragg *et al.*, 1965). In contrast, the effects of sorghum ergot on chickens was unknown, although the main alkaloid present, dihydroergosine (DHES), has been reported to be much less toxic to laboratory animals than rye ergot alkaloids (Frederickson *et al.*, 1991).

An ergot-rich sorghum fraction was separated from infected sorghum by floatation in 10% saline and air dried. This 'ergot-rich sorghum' contained about 120 g/kg by weight of ergot sclerotes and was incorporated into nutritionally similar diets at levels equivalent to 0, 12.5, 25 and 50 g ergot sclerotes/kg diet. Each diet was fed to six replicates of six broiler chicks of each sex from 3 to 23 days of age and the results are shown in the table.

Ergot sclerotes (g/kg):	0	12.5	25	50	SEM
Liveweight (g/bird)	737.8 ^a	735.8 ^{ab}	713.9 ^b	679.9 ^c	7.72
Food intake (g/bird)	950.9 ^a	951.3 ^a	948.1 ^a	896.7 ^b	10.18
FCR (g:g gain)	1.408 ^a	1.409 ^a	1.460 ^b	1.502 ^c	0.0113
GE of diet (MJ/kg DM)	20.1	19.9	19.7	19.4	
ME _n of diet (MJ/kg DM)	13.75 ^a	13.41 ^b	12.93 ^c	12.42 ^d	0.055
Diarrhoea at 8 days (%)	0.0 ^a	4.2 ^a	16.9 ^b	33.9 ^c	4.31
Diarrhoea at 14 days (%)	0.0 ^a	2.8 ^{ab}	8.9 ^b	16.4 ^c	2.45
Mortality at 20 days (%)	1.4 ^a	1.4 ^a	2.8 ^a	20.8 ^b	2.74

Means without a common superscript are significantly different at P<0.05.

The significant depression in growth with 25 g ergot/kg was accompanied by poorer FCR, a reduction in dietary ME_n and increased diarrhoea, but feed intake was unaffected. At 50 g ergot/kg an even greater depression in performance occurred, with reduced food intake and substantial mortality. In a few cases where death was observed, it was preceded by apparent gasping for breath. No effects on the feet, beak or comb were observed. These results are in agreement with reports that DHES in sorghum ergot produces less severe effects on the periferal circulation than rye ergot alkaloids. However, some toxic effect clearly remains at levels of 25 and 50 g ergot/kg, perhaps as a contribution from the more potent clavine alkaloids - agroclavine, chanoclavine and elymoclavine which are present as minor components. The role of these alkaloids needs more investigation.

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¹ Queensland Poultry Research and Development Centre, Alexandra Hills, Qld 4160.

² Queensland Dept of Primary Industries, Animal Research Institute, Yeerongpilly, Qld 4105.

RESTING AND DIET IN THE RECOVERY OF LAYING HENS FROM AN EGG PRODUCTION DROP ASSOCIATED WITH AVIAN ENCEPHALOMYELITIS

D. ROBINSON

The nutrition and husbandry of laying hens are known to affect their reaction to the presence of disease organisms. Increased dietary concentrations of various nutrients may be beneficial for layers subjected to stress resulting from a disease or vaccine challenge. Induced resting from lay is sometimes recommended as a means of accelerating recovery from egg production drops. This report summarises the effects of resting and diet following a severe decline in egg production and a marked deterioration in shell quality in a multi-strain flock commencing at 53 weeks of age. Diagnostic tests indicated that the production drop was caused by Avian Encephalomyelitis virus, although the persistent nature of the problem suggests that other factors may also have been involved. The affected flock comprised equal numbers of four strains of bird (two local and two imported) of the same age, and all strains were affected to a similar extent and responded similarly to the subsequent treatments. Prior to this study, birds of each strain were housed in randomly distributed groups of eight (four adjacent two-bird cages) in an open-sided shed, and more than half of the groups were affected. At 65 weeks of age, 144 groups of birds were selected on the basis of the previous four weeks' production records and divided into two equal-size categories, L and H, having average laying rates of approximately 27% and 74% respectively and equally representing all four strains. At this stage the mean body weight of the L birds was 0.221 kg lower than that of the H birds and their mean egg weight was 1.4g lower. At 66 weeks of age the two sets were divided into three sub-sets: unrested, rested by feeding barley for nine days (short rest) and rested by feeding barley for 18 days (long rest). Two post-rest diets were fed to an equal number of groups in each subset from the end of the rest inducement period until the end of the trial. These diets were low density - 11.0 MJ/kg ME and 15.4% protein, and high density - 12.1 MJ/kg ME and 18.9% protein. Data were analysed for the period from 72 weeks of age (which is the average age at which the long-rested birds returned to a peak rate of lay) to 81 weeks of age.

Compared with the no-rest treatment, short and long rests had little or no effect on egg number, egg weight or percentage of eggs with shell defects in the L birds, but increased the rate of lay by 8-14 percentage points and mean egg weight by 1.3g and reduced the proportion of eggs with defective shells by 10-13 percentage points in the H birds. Subsequent feed intake increased with increasing length of rest in both the L and H categories. While egg number and shell quality of the H birds was not significantly affected by dietary nutrient density, the rate of lay of the L birds was 20 percentage points higher and the proportion of shell defects was 20 percentage points lower with the high density diet than with the low density diet. The high density diet increased mean egg weight by 4.2g in the L birds and by 2.3g in the H birds. Compared with the low density diet, the high density diet resulted in lower feed intake in the H birds and higher feed intake, substantial bodyweight gains and lower mortality in the L birds.

It is concluded that birds in lay which suffer a severe set-back require a high quality diet and may not be rejuvenated by resting.

Queensland Poultry Research & Development Centre, PO Box 327, Cleveland, Ql 4163.

THE POST-HATCH DEVELOPMENT OF INTESTINAL ENZYME FUNCTION IN BROILER CHICKS

P.A. IJI and D.R. TIVEY

Broiler chicks are hatched with larger body and gastrointestinal mass than egg-type chicks but the specific activities of digestive enzymes, including amylase, lipase and trypsin have been observed to be lower in the former than in the latter (Nir *et al.*, 1993). Broiler chicken productivity might be further improved if the limitation in digestive function could be overcome through selection and breeding. To enable such improvement, there is a need to understand the natural development pattern of digestive function in important strains of broiler chickens utilized around the world.

In the present study, we monitored the development of four intestinal enzymes: sucrase (EC. 3.2.1.48), maltase (EC. 3.2.1.20), aminopeptidase N (APN; EC. 3.4.11.2) and alkaline phosphatase (AP; EC. 3.1.3.1) in the Steggles x Ross strain (Australia Poultry Pty Ltd.), developed in the early 1990s. The chicks were maintained on a commercial diet (Milling Industry Stockfeeds, Murray Bridge, South Australia). Seven chicks were randomly selected at hatch and thereafter, every 7th day until 21 days of age for assessment. The chicks were euthanized through intravenous administration of Lethobarb™ (pentobarbitone sodium). Intestinal samples were collected from the duodenum, jejunum and ileum and snap-frozen. Brush-border membrane vesicles were prepared and used to study the specific activity of membrane-bound digestive enzymes, using biochemical techniques. The spatial localization of these enzymes on the crypt:villus axis was also examined by histochemistry on sections obtained from fresh-frozen tissues.

The specific activity of sucrase, maltase, aminopeptidase APN and AP declined ($P < 0.001$) with age. The reduction in activity varied with intestinal segment; between hatch and 21 days of age, maximum reductions in activity were 69.8, 91.7, 73.9 and 83.3 % respectively for maltase, sucrase, APN and AP. There were also variations in the specific activities of these enzymes between intestinal sites. At hatch, the specific activities of maltase ($P < 0.001$) and sucrase ($P < 0.05$) were higher in the ileum than in the duodenum or jejunum. AP was expressed at a higher ($P < 0.01$) activity in the duodenum than in the jejunum or ileum both at hatch and by 21 days of age. The specific activities for the other enzymes at the three segments were similar at 21 days of age. Enzyme histochemistry revealed significant increases in the total activity of α -glucosidase (AG) ($P < 0.001$), APN and AP ($P < 0.05$) mainly as a result of increased surface area of the villus. Activity per unit villus surface area declined with age.

Enzyme profile analysis demonstrated the expression of activity throughout the crypt:villus axis although peak expression occurred on the mid-villus for AG and APN, and more towards the villus tip in the case of AP.

The results confirm an increase in total enzyme activity with age but the reduction in specific activities may indicate that digestive function lags behind the rapid growth rate reported by Iji and Tivey (1997) for this strain of broiler chickens.

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Dept of Animal Science, University of Adelaide, Waite Campus, Glen Osmond, S.A. 5064.

LOW PROTEIN DIETS FOR LAYERS : HOW LOW CAN WE GO?

D.J. FARRELL², D. ROBINSON¹ and J. PRIEST²

The concept of low protein diets in livestock production is attractive because they maximise the use of dietary protein and reduce nitrogen excretion. Our previous attempts to maximise layer performance on low protein diets were not successful. Here we formulated two diets on the basis of ingredient composition reported in the literature. Two other diets were formulated which maximised the use of crude protein in grains by balancing the protein with free amino acids and small amounts of a protein concentrate mixture or sunflower meal. Other amino acids were added where deficient, plus limestone, minerals and vitamins. Electrolytes were balanced here but not in previous experiments.

Major dietary ingredients in layer diets and results of bird performance.

Diet	1	2 ¹	3	4 ²	5	SEM
Major ingredient composition (g/kg)						
Sorghum			166			
Maize	264	570	250	630	Control	
Barley	264			116		
Wheat	264		261			
Soybean meal (45% CP)		90		115		
Mill run		206	100			
Sunflower meal (32% CP)			39			
Protein concentrate	81					
Tallow	15	4	6			
D-L methionine	2.3	2.1	2.6	2.5		
L-lysine	4.2		5.4	3.1		
Crude protein (g/kg)	129	139	119	122	170	
Layer performance (n = 25) over 12 weeks						
Egg no (%)	88.1	85.0	87.2	88.2	86.7	1.56
Egg weight (g)	53.5	54.6	53.5	53.3	53.8	0.55
Egg mass (g)	47.0	46.4	46.7	48.9	46.7	0.98
Food intake (g/d)	114.9	118.5	118.3	118.5	119.1	2.50
Feed efficiency (g/g)	2.46	2.60	2.56	2.45	2.57	0.070

¹ Diet 2 Summers (1993); ² Diet 5 Keshavarez and Jackson (1992).

There was no difference ($P>0.05$) in any production parameter measured. Although there were differences between months, there was no treatment by time interaction. The crude protein content of diets 3 and 4 was about 12% on an 'as is' basis compared to 17% on the control diet (5). Diet 3 contained only 4% sunflower meal as the protein concentrate. These results are encouraging and larger-scale and longer-term experiments are in progress.

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¹ Queensland Poultry Research and Development Centre, Alexandra Hills, Qld 4161.

² The Department of Agriculture, The University of Queensland, St Lucia, Qld 4072.