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# Effect of vegetable diets versus diets with processed animal proteins on performance and health status of laying hens

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ANIMAL SCIENCES GROUP
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**Abstract**
Meat and bone meal and meat meal in diets and their effects on performance and health status of laying hens has been reviewed. Properties of dietary animal proteins are compared with dietary vegetable proteins and possible causative factors for differences in performance and health are described in this report.



**Keywords**
Laying hen, meat and bone meal, meat meal, protein, amino acids, minerals, nutraceuticals, phytoestrogen, performance

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**Samenvatting**
Het gebruik van diermeel in voer en het effect ervan op technische resultaten en gezondheid bij leghennen is bestudeerd in een deskstudie. De eigenschappen van dierlijk eiwit zijn vergeleken met die van plantaardig eiwit en mogelijke oorzaken voor een verschil in performance en gezondheid worden beschreven in dit rapport.



**Trefwoorden:** leghen, diermeel, vlees- en beendermeel, vleesmeel, eiwit, aminozuren, mineralen, nutraceuticals, fyto-oestrogeen, technische resultaten.

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# Effect of vegetable diets versus diets with processed animal proteins on performance and health status of laying hens

# Effecten van plantaardig voer versus voer met dierlijke eiwitten op productieprestaties en gezondheidsstatus van leghennen

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October 2008

## Preface

The use of animal protein sources in animal diets was criticized after the occurrence of BSE in ruminants and Creutzfeld Jacob disease in humans. Since the ban on the use of animal proteins in poultry diets as of 2001, many poultry farmers are complaining about adverse effects on performance, egg quality and behaviour of laying hens. Laying hen farmers connect various problems like Infectious Bronchitis, Infectious Laryngotracheitis, coccidiosis, E. coli (also in vaccinated flocks), impaired feathering, hypodermic dermatitis, and chronic enteritis to the ban on animal proteins. Beside these effects, the higher worldwide demand for dietary protein sources and replacers such as soybean meal resulted in increased feed costs. At this moment the international community is discussing how to reintroduce animal proteins into poultry diets within the safety margins related to BSE.
Animal protein is an excellent feedstuff for laying hens because the amino acid balance meets the amino acid requirement of the laying hen. Animal protein is low in salts (e.g. potassium) which is beneficial to the quality of the manure and eggs. Animal protein is a common feedstuff for laying hens in a natural environment. Providing diets without animal proteins is in fact unnatural in laying hens.
The Product Board for Poultry and Eggs has ordered the Animal Sciences Group of Wageningen UR to conduct a desk study to determine the effects of vegetable diets versus diets with processed animal proteins on performance and health status of laying hens. The results are presented in this report.

Project leader
Marinus van Krimpen, PhD.
October 2008

## Summary

Since the ban on the use of animal proteins in poultry diets as of 2001, many poultry farmers are complaining about adverse effects on performance results, egg quality and behaviour. The results of a desk study on the effects of vegetable diets versus diets with processed animal proteins on performance, behaviour and health status of laying hens are presented in this report.

The most important conclusions of this study are:

- Processed animal proteins are rich in protein (lysine and methionine), calcium and phosphorus (supplied by the bone fraction), and vitamin $B_{12}$.
- Digestibility of amino acids and availability of phosphorus is higher in animal protein sources compared with vegetable protein sources. This results in a substantially lower excretion of nitrogen and phosphorus, and to a lower ammonia emission.
- The ratio of bone to soft tissue determines the ash content of processed animal proteins. A higher ratio of bone to soft tissue increases the ash content which has a negative effect on protein and energy concentrations in animal protein sources.
- The bone fraction contains collagen which is deficient in most essential amino acids (tryptophan, sulfur amino acids and isoleucine). An increase in bone content of the raw materials negatively affects protein quality due to the high collagen content and poor amino acid balance.
- The quality of animal protein sources (availability of amino acids) is affected by heat treatment and raw material composition.
- Increasing pressure during processing of meat and bone meal decreases the amino acid digestibility in laying hens.
- After the ban on the use of animal proteins, the amount of soybean meal, wheat and corn was increased significantly in diets without animal products. The use of phytase was also increased to make phytic acid in vegetable feedstuffs available. Monocalcium-phosphate was included in vegetable diets to increase the calcium and phosphorus content in order to meet the requirements of the laying hen.
- Higher inclusion rates of vegetable protein sources increase the proportion of fermentative degradable carbohydrates (oligosaccharides) in the diet which may result in a high fermentative activity in the gut and herewith adversely affecting gut health.
- In research it was shown that vegetable diets compared to animal protein-enriched diets resulted in a decreased egg production rate and an increased egg weight. Egg mass was not affected. Specific gravity of the eggs, Haugh Unit, percentage eggshell weight was decreased and the ratio of broken/cracked eggs was increased in laying hens fed diets without meat and bone meal.
- An effect of dietary protein source on the incidence of pecking damage in layers has not been demonstrated experimentally, however practical evidence suggests a higher incidence of feather pecking and chronic gastroenteritis at different ages after the ban on animal protein. More research in this field should be developed.
- It is suggested that any suppressive effect on feather pecking induced by animal protein is due to beneficial compounds only observed in these animal protein sources, for instance (the animal protein factor) vitamin $B_{12}$, or bioactive peptides. However, it is also conceivable that a detrimental compound in plant protein sources could increase feather pecking behavior, for instance phytoestrogens. More research in this field should be developed.

More research on the feeding value of pig meat meal in poultry diets should be considered as well as the effects of this feedstuff on behaviour and gastro intestinal health. Such experiments might help to promote the re-introduction of pig meat meal in laying hen diets.

## Samenvatting


Leghennenhouders klagen over verminderde productie en eierenkwaliteit en afwijkend gedrag bij de leghennen na het verbod op het gebruik van dierlijke eiwitten in pluimveevoer in 2001. De resultaten van een literatuurstudie naar het effect van een voeders zonder en met dierlijke eiwitten op productie, gedrag en gezondheid van leghennen worden gepresenteerd in dit rapport.

De meest belangrijke conclusies van deze studie zijn:

- Dierlijke eiwitbronnen zijn rijk aan eiwit (lysine en methionine), calcium en fosfor (geleverd door de botfractie) en vitamine $B_{12}$.
- De aminozuur verteerbaarheid en het gehalte aan beschikbaar fosfor is in dierlijke eiwitbronnen hoger dan in plantaardige eiwitbronnen. Dit leidt tot een lagere stikstof- en fosforuitscheiding en tot minder ammoniakemissie.
- De verhouding tussen bot en weefsel bepaalt het asgehalte in dierlijke eiwitbronnen. Een ruimere verhouding van bot ten opzichte van weefsel verhoogt het asgehalte en verlaagt het eiwit- en energiegehalte.
- De botfractie bevat collageen wat deficiënt is aan de meeste essentiële aminozuren (tryptofaan, zwavelhoudende aminozuren en isoleucine). Een toename van het botgehalte in de dierlijke eiwitbron heeft een negatief effect op de eiwitkwaliteit vanwege het hoge collageengehalte en een suboptimaal aminozuurprofiel.
- De kwaliteit van dierlijke eiwitbronnen (beschikbaarheid van aminozuren) wordt beïnvloed door de hittebehandeling tijdens processing en het uitgangsmateriaal.
- Een hogere druk tijdens processing van dierlijke eiwitbronnen vermindert de aminozuurverteerbaarheid bij leghennen.
- Na het verbod op het gebruik van dierlijke eiwitbronnen is de hoeveelheid sojaschroot, tarwe en maïs aanzienlijk toegenomen in de voeders. Het gebruik van fytase is ook toegenomen om fytinezuur beschikbaar te maken in plantaardige grondstoffen. Extra monocalciumfosfaat wordt toegevoegd aan voeders zonder dierlijke eiwitbronnen om de behoefte van leghennen aan calcium en fosfor te dekken.
- Het hogere aandeel van plantaardige eiwitbronnen vergroot het aandeel fermentatief afbreekbare koolhydraten (oligosacchariden) in het voer wat resulteert in een hoge fermentatieve activiteit in de darm en dit kan leiden tot een verminderde darmgezondheid.
- In onderzoek is aangetoond dat plantaardige eiwitbronnen in vergelijking met dierlijke eiwitbronnen resulteerden in een verminderde eiproductie en een toename van het eigewicht. De eimassa werd niet beïnvloed. Het soortelijk gewicht van de eieren, de dikwithoogte en het percentage eischaal nam af en het aandeel breukeieren nam toe bij leghennen die voeders verstrekt kregen zonder dierlijk eiwit.
- Onderzoeksresultaten wijzen niet op een eenduidig effect van de eiwitbron op pikkerij bij leghennen, alhoewel praktijkervaringen er op wijzen dat pikkerij en chronische darmontsteking op verschillende leeftijden meer voorkomen na het verbod op het gebruik van dierlijke eiwitbronnen. Meer onderzoek op dit gebied is gewenst.
- In de literatuur wordt gesuggereerd dat dierlijke eiwitbronnen mogelijk pikkerij kunnen onderdrukken als gevolg van bepaalde gunstige stoffen die alleen aanwezig zijn in dierlijke eiwitbronnen zoals bijvoorbeeld de zogenoemde ‘dierlijke eiwitfactor’ (vitamine $B_{12}$) of bioactieve peptiden. Echter, het kan ook zo zijn dat in plantaardige eiwitbronnen ongunstige stoffen aanwezig zijn die de pikkerij juist triggeren zoals bijvoorbeeld fyto-oestrogenen. Ook op dit gebied is meer onderzoek gewenst.

Overwogen kan worden om meer onderzoek naar de voederwaarde van varkensvleesmeel in leghennenvoer uit te voeren en daarin het effect van deze grondstof op het gedrag en darmgezondheid te betrekken. Deze experimenten kunnen bijdragen aan een snellere herintroductie van varkensvleesmeel in leghennenvoer.

# Contents

# 1 Introduction

Since the ban on the use of animal proteins in poultry diets as of 2001, many poultry farmers are complaining about adverse effects on performance, egg quality and behaviour of laying hens. From that time also problems with chicken mites increased and hens were housed more and more in alternative housing systems. Laying hen farmers however, connect various problems like Infectious Bronchitis, Infectious Laryngotracheitis, E. coli (also in vaccinated flocks), impaired feathering, hypodermic dermatitis, and chronic enteritis to the ban on animal proteins. There is no clear evidence that these adverse effects are primarily caused by the ban on the use of animal proteins but farmers see a link between these effects and the ban.
Beside these effects, the higher worldwide demand for dietary protein sources and replacers such as soybean meal resulted in increased feed costs. At this moment the international community is discussing how to reintroduce animal proteins into poultry diets within the safety margins related to BSE.

The objective of the desk study was to study the added value of animal proteins in relation to vegetable proteins in diets on performance, health and behaviour of laying hens.

The results of a desk study on the effects of vegetable diets versus diets with processed animal proteins on performance, behaviour and health status of laying hens are presented in this report. Chapter 1 provides a historical overview on the use of animal protein in animal feeds. In Chapter 2, the chemical composition and effects of processing on the quality of meat and bone meal are described. In Chapter 3 the ban on animal protein and the consequences for diet composition is presented. Performance results and effects on behaviour and health in laying hens fed vegetable or animal protein diets are discussed in Chapter 4. Finally, the possible mode of action of processed animal protein on performance and health status is discussed in Chapter 5.

# 2 Historical perspective

The use of animal protein in poultry feed has been reported to occur in the late 1880's prior to the establishment of a rendering industry. By-products from country slaughterhouses and on farm butchering were sold for chicken feed. Scott et al. (1969) wrote: "Early experience has demonstrated protein sources such as fish meal, meat scraps and dried skim milk, when added to poultry diets, produced results which were vastly superior to those obtained with similar diets containing only plant proteins". This observation implies that animal proteins have been a very integral part of the increase of overall efficiency in poultry production. Scott et al. (1969) reported: "that during World War II, because of the known nutritional value of liver in human diets, there was an effort to eliminate expensive and scarce animal proteins from animal diets. Subsequently, the use of vegetable protein diets reduced the rate of growth in chickens and pigs and decreased the hatchability of eggs". Presented responsible factors for the early superiority of animal proteins as compared with plant proteins were 1) the calcium and phosphorus supplied by the bone fraction in animal protein supplements; 2) B-complex vitamins, particularly riboflavin, in dried skimmed milk and whey; 3) vitamin $B_{12}$, which is present in all animal materials, but not in plants; and finally 4) the amino acids methionine and lysine, which are present in the protein fraction of fish, eggs and milk at much higher levels than in the common protein supplements of plant origin (Scott et al., 1969). By 1950, identification of vitamin $B_{12}$ as the animal protein factor, and its commercial synthesis, made it possible to develop diets without animal protein for non-ruminants (Haugen et al., 1985). The corn-soybean meal diet became more popular, and since then meat meal had to compete in the market with soybean meal and other plant protein sources (Denton et al., 2005).

Nutritionists have utilized meat and bone meal in poultry diets for many years (Kraybill, 1928; Prange et al., 1927, 1928a,b; Kratzer and Davis, 1959; Skurray, 1974; Waldroup and Adams, 1994; Dale, 1997; Parsons et al., 1997). The primary advantages associated with the utilization of meat and bone meal in poultry diets have been the high digestibility of amino acids in meat and bone meal as well as the biological availability of phosphorus in animal protein (Waldroup and Adams, 1994; Parsons, 1996, 1998; Sell and Jeffrey, 1996).

After the ruminant to ruminant ban (1989 in the Netherlands, 1994 EU-wide) inclusions of meat and bone meal in poultry rations increased because of favourable supply and pricing. The relatively high inclusion level of meat and bone meal in poultry diets remained until the total ban on meat and bone meal in 2001 (Table 1).

**Table 1** Historical overview of measures to avoid spread of BSE (Bovine Spongiforme Encephalopathy) in the Netherlands

| Date | Measure |
|---|---|
| January 1989 | Ban on processing meat and bone meal from ruminants in feeds for ruminants. |
| September 1990 | Import ban on meat and bone meal from ruminants originated from UK. |
| January 1993 | The Dutch animal feed industry establishes measures to avoid cross contamination at the production of compound feeds. Cross contamination may occur when compound feeds for different species are produced within the same production unit. The measure includes the prohibition of producing compound feeds for ruminants in the same production unit as where feeds are produced that includes more than 6% meat and bon |
| August 1994 | European ban to include meat and bone meal originated from ruminants in compound feeds for ruminants. |
| June 1997 | Legal obligation to sterilize raw materials used for meat and bone meal during 20 minutes at a temperature of 133 °C and a pressure of 3 bar with a maximum particle size of 50 mm. |
| August 1997 | Some tissues will be categorized as high risk material because those tissues will increase the risk for spread of BSE. High risk material will be destroyed by waste incineration. |
| March 1999 | Ban on the production of compound feeds for ruminants that contain animal proteins. For practice this implies a zero tolerance and compound feeds for ruminants may only be produced in separate production units or in separate production locations. |
| January 2001 | European ban on the use of animal proteins in all feeds for farm animals. |
| May 2003 | Regulation animal by-products which categorizes animal by-products in different classes. Category 1: High risk material; Category 2: Medium risk material; Category 3: Low risk material. Only Category 3 material may be used in animal feed. This material originates from healthy animals which may be used for human consumption (skin, hoof, feathers, blood of non-ruminants). |

# 3 Specifications of the quality of meat and bone meal

## 3.1 Chemical composition

In our modern society not all processed animal material is used for human consumption. This material, mainly existing of fat, meat, organs, bones, blood and feathers is transported from processing plants to rendering plants and is further processed under regulatory requirements into a powdery substance: meat meal or meat and bone meal. These meals can be incorporated in animal feed as a protein source. Protein levels of meal are determined by the meals' crude protein content, sorted into three main categories: 50%, 48% and 45%. Meat and bone meals with a protein content of less than 45% are likely to contain too much bone material to be suitable for animal feed. Ravindran and Blair (1993) stated that meat meals may contain 20-60% bone. Depending on the ratio of bone to soft tissue used in processing, the finished product is designated as 'meat meal' (containing >55% crude protein and <4.4% phosphorus) or 'meat and bone meal' (containing <55% crude protein and >4.4% phosphorus).

Nutritional quality of commercial meat meal is also extremely dependant on the type of raw material rendered. One of the constituents that can affect the composition and protein quality of meat and bone meal is the bone fraction. For example, increased bone or ash content has been shown by Dale (1997), Mendez and Dale (1998), Wang and Parsons (1998a) to have a negative effect on protein and energy concentrations.

Much of the variation in the nutritive value of meals is due to the level and availability of essential amino acids. Pepsin digestible protein, the amount of crude protein solubilised in vitro under standard conditions and time, is a minimum quality standard which indicates the nutritional value of meat and bone meal. The protein keratin in wool, feathers and hair is not digestible by pepsin unless the rendering process includes a pressure cycle capable of hydrolysing keratin. Meat and bone meals with low pepsin digestibility are likely to contain a high proportion of wool, hair or feathers. Non-hydrolysed protein from these sources has no nutritional value for pigs and poultry. Another specification which affects the nutritive value of meat and bone meal is ash. Ash is the residue remaining after incineration under the conditions specified for the test, and indicates the amount of minerals in the meal. Bone is the main source of ash. The bone content of meat meal provides calcium and phosphorus, thus helping to supply necessary minerals to the diet. The effects of ash on protein quality are not totally clear. It is expected that some decrease in protein quality with increased ash will occur due to the changes in AA concentrations. In addition, an increase in ash could further decrease protein quality if digestibility of AA is reduced. Johnson and Parsons (1997) and Johnson et al. (1998) showed that protein efficiency ratio (PER; chick weight gain per unit of CP intake) decreased from 1.7 to 1.0 as ash content increased from 24 to 35% in two samples of meat and bone meal. In contrast, mean AA digestibility of the 24 and 35% ash meat and bone meal samples were not significantly different ($P > 0.05$) (70.8 and 76.3%, respectively). Thus, increased bone ash had no negative effect on AA digestibility in the two meat and bone meal samples evaluated in the previous studies. Shirley and Parsons (2001) concluded that the reduction in protein quality of meat and bone meal as ash content increases is almost entirely due to negative effects on AA balance or profile of the meat and bone meal, and not because of reduced AA digestibility.

The maximum moisture content of meal is specified as 10%. Meals with higher moisture levels can support mould growth and may not be free-flowing. High moisture levels also dilute the nutrients in meat and bone meal. Fat in meal is useful in that it contributes energy to a ration. As with moisture content, however, high fat levels can cause meal to cake in bins and chutes. Meat meal colour is usually light brown to dark brown. Lighter coloured meals generally come from raw materials that contain a high proportion of bone, with the light colour due to the white particles of bone in the meal. Dark coloured meals are also influenced by raw materials—for example, if raw materials contain blood or if blood is added to the raw material or finished meal, the meal will have a darker colour. Raw materials that contain unwashed gut materials tend to be dark in colour because of the pigmentation contributed by the gut contents. Meat meal colour is also affected by the temperature applied during rendering, with excessive temperature producing a dark meal. The particle size of meals is specified by feed millers because a meal which is not ground uniformly is difficult to mix with other ingredients and cannot be formed into pellets satisfactorily.
The chemical composition of meat and bone meal is rather variable depending on the used raw materials during production. The origin (ruminants, pig, poultry) of meat and bone meal is not presented in most papers. Feed value tables distinguish meat and bone meal with different classes of protein or fat content. Ash content of meat and bone meal and, consequently, mineral content may also vary considerably depending on the raw material.

To get an idea of differences in composition between meat meal and bone meal, average composition of animal meal and bone meal based on a large number of samples are given in Table 2.

**Table 2** Overall averages in meat meal and bone meal (g/kg) (Verband Fleischmehlindustrie, 2001)

| | Meat meal | Bone meal |
|---|---|---|
| Crude ash | 210 | 395 |
| Crude protein | 550 | 450 |
| Ether extract | 120 | 100 |
| Gross energy (MJ/kg) | 18.5 | 12.6 |
| Phosphorus | 31 | 61 |
| Calcium | 60 | 120 |
| Lysine | 31.0 | 22.4 |
| Methionine | 7.4 | 5.2 |
| Threonine | 20.0 | 12.8 |
| Tryptophan | 5.0 | 2.0 |

Marked variations were observed among samples of fish meal, meat and bone meal and meat meal, highlighting significant batch-to-batch differences. (Karakas et al., 2001) studied the nutritive value of the meat and bone meals originated from cattle or pigs in broiler diets at 4 weeks of age. The $AME_n$ content of the meat and bone meal in this study ranged from 2,511 to 3,115 kcal/kg (10.51 to 13.04 MJ/kg) DM. In the experiment, as was expected, the $AME_n$ content of meat and bone meal (10% inclusion level) decreased significantly at a higher ash content. The $AME_n$ contents were higher in meat and bone meal from pig origin than in meat and bone meal from cattle origin (Table 3). This difference seems reasonable, considering the lower percentage of bone in the pig meal samples. The results suggest a minor effect of the ash concentration on amino acid digestibility.

**Table 3** $AME_n$, excretal digestibility of crude protein and total amino acids of diets with an inclusion level of 10% cattle and pig meat and bone meal, varying in ash content (g/kg; Karakas et al., 2001)

| | $AME_n$ Kcal/kg DM | Crude Protein Digestibility % | Total AA Digestibility % |
|---|---|---|---|
| Basal diet | 3488 | 85.0 | 87.7 |
| MBM cattle 10%, ash 200 | 2857 | 64.8 | 62.2 |
| MBM cattle 10% ash 300 | 2955 | 64.1 | 63.3 |
| MBM cattle 10% ash 400 | 2661 | 65.6 | 60.1 |
| MBM pigs 10% ash 200 | 3115 | 64.3 | 61.7 |
| MBM pigs 10% ash 300 | 3024 | 66.5 | 64.8 |
| MBM pigs 10% ash 400 | 2511 | 66.4 | 61.4 |

Huang et al. (2007) studied the apparent ileal digestibility of amino acids in feed ingredients in broilers and layers (42 days and 60 weeks of age, respectively). The results for meat and bone meal are presented in Table 4. The digestibility of crude protein and the average digestibility of amino acids in meat and bone meal were similar for broilers and layers, except for that of histidine, glycine, alanine and tyrosine which was higher in layers.

**Table 4** Apparent ileal digestibility coefficients of crude protein and amino acids in meat and bone meal for broilers and layers (Huang et al., 2007)

| Parameters | Broilers | Layers |
|---|---|---|
| Crude protein (N x 6.25) | 0.75 | 0.76 |
| Essential amino acids | | |
| Threonine | 0.76 | 0.75 |
| Valine | 0.80 | 0.81 |
| Methionine | 0.84 | 0.84 |
| Isoleucine | 0.81 | 0.82 |
| Leucine | 0.82 | 0.83 |
| Phenylalanine | 0.81 | 0.83 |
| Histidine | 0.78 | 0.80 |
| Lysine | 0.81 | 0.81 |
| Arginine | 0.82 | 0.83 |
| Non-essential amino acids | | |
| Aspartic acid | 0.66 | 0.67 |
| Serine | 0.73 | 0.74 |
| Glutamic acid | 0.79 | 0.79 |
| Glycine | 0.75 | 0.80 |
| Alanine | 0.79 | 0.82 |
| Tyrosine | 0.81 | 0.83 |
| Average | 0.79 | 0.80 |

Ravindran and Blair (1993) compared the chemical composition and amino acid profiles of soybean meal, as the major vegetable feedstuff to substitute meat and bone meal in poultry diets, with some commonly available animal protein supplements (Table 5).

**Table 5** Average chemical composition and amino acid profiles of soybean meal and some commonly available animal protein supplements (Ravindran and Blair, 1993)

| | Soybean meal | Fishmeal menhaden | Meat meal | Meat and bone meal | Blood meal | Skim milk powder |
|---|---|---|---|---|---|---|
| Chemical composition (% DM) | | | | | | |
| Crude protein | 45.0 | 62.0 | 60.0 | 45.0 | 80.0 | 32.0 |
| Crude fat | 2.0 | 10.0 | 9.0 | 8.0 | 1.0 | 1.0 |
| Crude fibre | 5.0 | 1.0 | 2.0 | 2.0 | 1.0 | 0.0 |
| Ash | 6.0 | 20.0 | 21.0 | 37.0 | 4.0 | 8.0 |
| Calcium | 0.2 | 5.0 | 6.0 | 11.0 | 0.3 | 1.2 |
| Phosphorus | 0.6 | 3.0 | 3.0 | 6.0 | 0.2 | 1.0 |
| Metabolizable energy (kcal/kg DM) | 2400 | 2850 | 2650 | 1700 | 700 | 2500 |
| Amino acid composition (g/100 g protein) | | | | | | |
| Arginine | 8.3 | 5.2 | 6.0 | 5.9 | 2.8 | 3.4 |
| Cystine | 1.0 | 1.0 | 0.8 | 0.6 | 1.6 | 1.3 |
| Glycine | 4.5 | 6.2 | 8.2 | 14.3 | 5.3 | 3.6 |
| Histidine | 3.0 | 2.3 | 3.2 | 3.3 | 3.6 | 2.6 |
| Isoleucine | 5.6 | 4.5 | 3.2 | 3.7 | 1.0 | 6.5 |
| Leucine | 8.2 | 8.0 | 8.5 | 6.4 | 12.4 | 10.2 |
| Lysine | 6.8 | 7.7 | 6.3 | 4.8 | 8.4 | 8.1 |
| Methionine | 1.4 | 2.9 | 1.3 | 1.2 | 1.2 | 3.1 |
| Phenylalanine | 4.9 | 3.6 | 4.5 | 4.0 | 6.0 | 4.9 |
| Threonine | 4.0 | 3.7 | 4.0 | 4.0 | 4.6 | 5.4 |
| Tryptophan | 1.4 | 1.2 | 1.1 | 0.7 | 1.2 | 1.4 |
| Tyrosine | 4.2 | 3.0 | 1.8 | 1.7 | 3.2 | 4.5 |
| Valine | 5.5 | 5.4 | 7.0 | 5.3 | 6.0 | 7.4 |

It is noticeable that animal meal contains a higher protein content but its amino acid availability is lower than in soybean meal. Metabolizable energy is higher due to the higher protein and fat content in meat meal. With respect to the amino acids relative to the protein content, meat meal contains less arginine, more glycine, less isoleucine, less tryptophan, less tyrosine and more valine compared with soybean meal.
Tryptophan is often the third limiting amino acid in poultry diets, and especially when diets are based on maize and meat and bone meal tryptophan content should be taken into account.
Ravindran et al. (2006) studied the total and ileal digestible tryptophan contents of feedstuffs for broiler chickens. They observed that the tryptophan digestibility coefficient in fish meal (0.77) and blood meal (0.84) was substantially higher than in meat meal (0.62), meat and bone meal (0.63) and feather meal (0.52).

## 3.2 Effects of processing on the quality of meat and bone meal

The quality of meals is affected by heat treatment and raw material composition. During rendering or drying, three types of nutritional damage may take place depending on time and temperature conditions:

1. Total destruction of amino acids
2. Maillard or browning reactions. For example, the free amino group of lysine reacts with certain types of sugars (carbohydrates, glycogen and blood-sugars) preventing the subsequent breakdown by enzymes in the gut.
3. Cross linking between amino acids, preventing the breakdown of the protein by enzymes.

Nutritional damage reduces both the availability of amino acids and the quality of the meal. Lysine is besides methionine and cysteine the first amino acid which becomes limiting for growth and, because it has a free amino group, it is often damaged during heat processing. As a result, the amount of available lysine in meal is used to provide an indicator of the degree of damage caused to the amino acids during meal production.
Meat meal derived from boning room cooking materials are of poorer nutritional quality than those derived from cooking materials from the slaughter floor. The protein keratin is a major constituent of feathers, wool and hair – materials that are high in crude protein. Unless the keratin is fully hydrolysed under pressure, it is not digested by pigs and poultry. Hard offal – bones, heads, hooves, skin cartilage and connective tissue contain a high collagen content. Eastoe and Long (1960) estimated that 83% of the protein in bone is collagen. Boomgaardt and Baker (1972) and Berdanier (1998) have shown that collagen and gelatin (refined collagen) are deficient in most essential amino acids such as tryptophan, sulfur amino acids, and isoleucine, while they are surfeit in hydroxyproline, proline, and glycine. Therefore, any increase in bone content of the raw materials may have a negative effect on protein quality due to its high collagen content and poor amino acid balance. In comparison, soft offal – muscle, gut and stomach – contain much higher levels of essential amino acids and will produce meals of greater nutritional value.
Shirley and Parsons (2000) studied the effect of pressure processing on amino acid digestibility of meat and bone meal for poultry. Three experiments evaluated the effects of different processing pressures on the digestibility of amino acid in meat and bone meal when the pressure processing was done after typical rendering (Experiments 1 and 2) or during the initial rendering process of raw materials (Experiment 3). In experiment 1, seven meat and bone meal samples from one large batch of commercially rendered beef meat and bone meal were processed at various pressures, temperatures, and times in an experimental batch cooker. The processing treatments were as follows: 0 psi (94 °C) for 20 min, 15 psi (121 °C; 103 kPa) for 20 min, 15 psi (121 °C; 103 kPa) for 30 min, 30 psi (133 °C; 207 kPa) for 20 min, 30 psi (133 °C; 207 kPa) for 30 min, 45 psi (147 °C; 310 kPa) for 20 min, and 60 psi (144 °C; 413 kPa) for 20 min. The first treatment consisted of cooking the meat and bone meal for 20 min with no pressure to determine if heat processing alone would have any effect on amino acid digestibility. In Experiment 2, commercially rendered beef meat and bone meal was processed in a commercial feather meal cooker at 45 psi (145 °C; 310 kPa) for 20 min or 60 psi (152 °C; 413 kPa) for 20 min. In Experiment 3, raw materials from a beef slaughter plant were pressure processed in an experimental cooker at 30 psi (207 kPa) for 20 min or 60 psi (413 kPa) for 20 min. No processing temperature data were provided in Experiment 3. Thus, experiments 1 and 2 differed from experiment 3 in that the pressure processing in experiments 1 and 2 was done on meat and bone meal that had already been commercially rendered (as done in the European
Union), whereas in Experiment 3, pressure processing was done during the initial rendering or cooking of the raw materials. Increasing pressure during processing reduced meat and bone meal cysteine-concentrations in Experiments 1 and 2. True digestibility of most amino acids were significantly decreased by increasing pressures in Experiments 1 and 2, and reductions were generally largest for cysteine and lysine, and increased with severity as pressure increased. For example, in Experiment 1, cysteine digestibility decreased from 65 to 50 to 15%, and lysine digestibility decreased from 76 to 68 to 41% as the meat and bone meal was processed at 0, 30, and 60

psi, respectively, for 20 min (Table 6). When the pressure processing occurred during the initial rendering of the meat and bone meal raw material (Experiment 3), a significant reduction in digestibility of most amino acids was observed only at 60 psi, and the decrease was much less than that observed in Experiments 1 and 2. These results indicate that pressure processing of meat and bone meal decreases the digestibility of amino acids for poultry. Since 1997, there is an EU legal obligation to sterilize raw materials used for meat and bone meal during 20 minutes at a temperature of 133 °C and a pressure of 3 bar with a maximum particle size of 50 mm. With relation to Table 6, 3 bar corresponds with 300 kPa. The column which corresponds with EU regulation on pressure is marked grey.

**Table 6** True amino acid digestibility coefficients for meat and bone meals processed under different pressures; Experiment 1 ((Shirley and Parsons, 2001))[1]

| Amino acid | Processing treatment (pressure/time)[2] | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | 0/0 | 0/20 | 15/20 | 15/30 | 30/20 | 30/30 | 45/20 | 60/20 | Pooled SEM |
| | | | | | (%) | | | | |
| Asp | 59.3a | 55.0ab | 49.0bc | 46.0bcd | 45.9bcd | 36.0e | 40.9cde | 26.3f | 3.2 |
| Thr | 80.7a | 78.3ab | 76.0abc | 70.9cd | 76.1abc | 67.5d | 73.1bcd | 54.5e | 2.2 |
| Ser | 83.2a | 79.4ab | 77.7abc | 69.4d | 78.4abc | 72.3cd | 77.5abc | 55.0e | 2.2 |
| Glu | 77.7a | 76.2ab | 72.4b | 71.0b | 74.1ab | 65.5c | 71.7b | 55.8d | 1.8 |
| Pro | 76.1a | 72.0ab | 68.4bcd | 69.4abcd | 71.7ab | 63.0de | 70.0abc | 60.3e | 2.3 |
| Ala | 79.1a | 76.8ab | 74.5b | 73.9bc | 76.2ab | 69.6c | 73.9bc | 61.4d | 1.6 |
| Val | 76.6a | 74.3ab | 70.7b | 72.6ab | 75.0ab | 64.6c | 70.6c | 56.6d | 1.8 |
| Ile | 78.7a | 76.4ab | 72.7b | 74.6ab | 76.3ab | 67.1cd | 72.0bc | 56.9e | 1.7 |
| Leu | 82.4a | 80.4ab | 77.3b | 76.9b | 79.5ab | 71.9c | 76.6b | 63.6d | 1.4 |
| Tyr | 84.2a | 77.7ab | 76.7abc | 73.8bc | 78.2ab | 69.4c | 76.5abc | 53.2d | 2.7 |
| Phe | 89.2a | 86.9ab | 84.5b | 83.7bc | 85.9ab | 79.4cd | 83.8bc | 69.1e | 1.6 |
| His | 78.0a | 75.5ab | 70.7b | 73.2ab | 72.1b | 65.0cd | 69.9bc | 52.8e | 1.9 |
| Lys | 75.5a | 72.1ab | 67.3bc | 66.1c | 67.6bc | 54.4d | 62.4c | 41.3e | 2.0 |
| Arg | 85.3a | 84.3ab | 83.4abc | 82.8abc | 85.6a | 80.4c | 84.6ab | 73.3d | 1.3 |
| Met | 80.6a | 76.5ab | 76.3ab | 74.0bc | 76.5ab | 69.6cd | 74.7bc | 62.3e | 1.8 |
| Cys | 64.8a | 56.8ab | 48.1bcd | 39.8de | 50.1bc | 30.1f | 45.5cd | 14.8g | 3.2 |
| Trp[3] | 72.0 | NM[4] | 65.0 | NM | 65.1 | NM | 60.6 | 55.4 | ... |

a–gMeans within a row with no common superscripts are significantly different ($P < 0.05$).

[1]Mean values of four cecectomized roosters per sample.

[2]The first number indicates the processing pressure in pounds per square inch (gauge). and the second number indicates the length of processing time in min. The 15, 30, 45, and 60 pounds per square inch correspond to 103, 207, 310, and 413 kPa, respectively.

[3]Trp values are calculated from one pooled excreta sample of four cecectomized roosters.

[4]NM = Not measured.

Meat meals are mainly produced from dry rendering plants. In the continuous wet rendering process, the wet rendered material is dried as a separate step, following centrifuging. Two types of dryers are available: contact dryers and pneumatic dryers. These may also be used for drying blood. Contact dryers are indirectly heated whereas pneumatic dryers can be directly fired. Due to rendering the wide variety of raw materials, differences in the final chemical composition of meat and bone meal can occur.

A loss of available lysine takes place during the initial stages of the batch cooking process due to Maillard reactions. Further damage only may occur as a result of quite serious deviations from normal dry rendering processing conditions (atmospheric pressure) involving temperatures of 140 °C or higher for longer than two hours. Similarly, pressure applied at any time during the processing cycle – to achieve, for example, hydrolysing of wool and hair – results in a substantial increase in the temperature of the contents with consequent damage to nutritional quality. The degree of destruction of amino acids in meals, caused by pressure and temperature, is estimated by feeding trials. The result of these may vary, depending upon the species of animal used to evaluate the meal. The results of chicken feeding trials indicate there is little difference between the availability of amino acids in meals made from similar raw materials which were either continuously wet rendered (and dried in a hot gas dryer at atmospheric pressure) or dry rendered (without pressure treatment, to normal endpoint of about 120 °C). However, pig feeding trials demonstrated that greater damage is caused by dry rendering.

# 4 Ban on animal protein in animal feeds and adjustments in diet composition after the ban

## 4.1 Ban on meat and bone meal

The aim of the ban on the use of animal protein in animal feeds is to eliminate BSE by removing the most important source of infection (infected animal protein). The measures included a ban on the feeding of processed animal proteins to animals which are kept, fattened or bred for the production of food (Table 7).

**Table 7** Application of feed controls in the Transmissible Spongiform Encephalopathies (TSE) Regulations 2006 (Anonymous, 2006)

| Protein source | Ruminants | Non-ruminant farmed animals |
|---|---|---|
| **Permitted animal proteins** - Milk, milk-based products and colostrum, Eggs & egg products, Gelatine from non-ruminants, Hydrolysed proteins derived from non-ruminants or from ruminant hides and skins | **Permitted** – subject to required sourcing and processing standards under Animal By-Product controls | **Permitted** – subject to required sourcing and processing standards under Animal By-Product controls |
| **Prohibited processed animal protein** (includes mammalian meat and bonemeal, meat meal, bone meal, hoof meal, horn meal, greaves, poultry meal, poultry offal meal, feather meal); Gelatine from ruminants | **Banned** (In addition to the restricted proteins listed below, and any animal protein not on the permitted list above) | **Banned** (Unprocessed animal by-products are also banned from feeding to farmed animals under Animal By-Product controls) |
| **Restricted proteins** (i.e. restricted to non-ruminant feed use) Fishmeal; Blood products; Blood meal (only to be fed to farmed fish); Di-calcium phosphate and tri-calcium phosphate (of animal origin only – not mineral) | **Banned** | **Permitted** – subject to authorisation to make feed with these products or registration to use it in complete feed on farms where ruminants are present |

## 4.2 Adjustments in diet composition after the ban

Pos (2001) compared standard diet compositions for laying hens just before and after the ban on animal proteins (Table 8).

**Table 8** Composition (%) of a standard laying hen diet with and without animal products, adapted from VVM, 2000, 2001 (Pos, 2001)

| Feedstuff | With animal products[1] | Without animal products[2] |
|---|---|---|
| Meat meal Crude fat > 120 | 3.161 | - |
| Feather meal | 2.000 | - |
| Animal fat | 4.000 | 4.000 |
| Peas CP < 220 | 5.000 | - |
| Grass meal CP 160-200 | 1.993 | - |
| Lucerne meal CP 160-180 | - | 1.186 |
| Corn | 25.000 | 30.000 |
| Corn gluten meal | 4.056 | 4.501 |
| Soybean (fullfat: toasted) | 0.023 | - |
| Soybean meal Crude fibre < 35 | 7.128 | 17.057 |
| Wheat | 24.772 | 28.824 |
| Wheat middlings | 12.500 | 0.190 |
| Sunflower seed meal CP 290 | - | 3.119 |
| Monocalcium-phosphate | - | 0.508 |
| Limestone | 8.835 | 9.066 |
| Salt | 0.185 | 0.27 |
| Premix 1% | 1.000 | 1.000 |
| Phytase, layers | 0.135 | 0.150 |
| DL-Methionine | 0.124 | - |
| L-Lysine HCL | 0.089 | 0.129 |
| | | |
| Calculated costs (€) | 14.42 | 14.85 |

[1]VVM, 6 November, 2000

[2]VVM, 9 February 2001

Meat meal and feather meal were used in standard diet compositions for laying hens before the ban. In total these animal protein sources comprised about 5% of the diet. In diets without animal protein sources, protein has to be supplied from vegetable protein sources. Which vegetable protein sources will be used in the diets depends on the price. The diet compositions before and after the ban in Table 8 are just an example. The diet composition may change due to changes in raw material prices.

The amount of soybean meal was increased significantly (10%) in diets without animal products. Also inclusion of corn and wheat was increased (5%) in diets without animal products. Phytase was increased relatively with 11% because of the presence of phytate-phosphorus in vegetable feedstuffs. Another clear difference is the lack of peas and the low content of wheat middlings in the composition after the ban. Sunflower seed meal extracted (3%) was included in the diet after the ban. Free Lysine was slightly increased while free methionine was not included anymore. After the ban monocalcium-phosphate was included to increase the calcium and phosphorus content in order to meet the requirements of the laying hen.

### *4.2.1 Protein*

The contribution of protein-rich feedstuffs to the crude protein content, digestible lysine and digestible methionine in laying hen diets before and after the ban on meat meal is presented in Table 9.

**Table 9** Relative contribution to Crude protein (CP), digestible lysine (Dig. Lys) and digestible Methionine (dig. Met) of feedstuffs in laying hen diets with and without animal products (Pos, 2001)

| Feedstuff | CP content (%)[1] | Relative share in total CP (%) | | Dig. Lys content (%)[1] | Relative share in total dig. Lys (%) | | Dig. Met content (%)[1] | Relative share in total dig. Met (%) | |
|---|---|---|---|---|---|---|---|---|---|
| | | With animal products | Without animal products | | With animal products | Without animal products | | With animal products | Without animal products |
| Meat meal | 58.3 | 10.6 | - | 2.68 | 12.9 | - | 0.68 | 5.9 | - |
| Feather meal | 82.4 | 9.4 | - | 1.59 | 4.9 | - | 0.45 | 2.5 | - |
| Peas | 21.2 | 6.1 | - | 1.25 | 9.5 | - | 0.18 | 2.5 | - |
| Grass meal | 17.7 | 2.0 | - | 1.47 | 1.2 | - | 0.16 | 0.9 | - |
| Lucerne meal | 19.6 | - | 1.1 | 0.44 | - | 0.8 | 0.15 | - | 0.4 |
| Corn | 8.5 | 12.2 | 14.6 | 0.15 | 5.7 | 7.2 | 0.16 | 11.0 | 12.0 |
| Corn gluten meal | 59.5 | 13.8 | 15.3 | 0.96 | 5.9 | 6.9 | 1.36 | 15.1 | 15.3 |
| Soybean | 35.6 | 0.05 | - | 1.88 | 0.066 | - | 0.42 | 0.0 | - |
| Soybean meal | 46.7 | 19.1 | 45.5 | 2.55 | 27.7 | 69.2 | 0.57 | 11.1 | 24.3 |
| Wheat | 11.1 | 15.8 | 18.3 | 0.26 | 9.8 | 11.9 | 0.16 | 10.9 | 11.5 |
| Wheat middlings | 15.4 | 11.0 | 0.2 | 0.45 | 8.6 | 0.1 | 0.18 | 6.2 | 0.1 |
| Sunflower seed meal | 28.7 | - | 5.1 | 0.78 | - | 3.9 | 0.54 | - | 4.2 |
| L-Lysine-HCl | - | - | - | 100 | 13.6 | - | - | - | - |
| DL-methionine | - | - | - | - | - | - | 100.0 | 34.0 | 32.2 |
| | | 100.0 | 100.0 | | 100.0 | 100.0 | | 100.0 | 100.0 |

[1] Source: CVB, 2000.

The animal protein sources meat meal and feather meal together supply 20% of the total crude protein. Meat meal, soybean meal and free lysine are the main sources of digestible lysine. Meat meal and feather meal together deliver 17.8% of the digestible lysine. The main source of digestible methionine in the diet with animal products is corn gluten meal. Meat meal and feather meal together deliver 8.4% of the total digestible methionine. In the feed composition methionine is almost completely supplied by soybean meal. In this composition soybean meal is therefore the main source of digestible methionine.

The soybean meal content increased most significantly after the ban on animal products. The relative contribution of soybean meal in CP and digestible lysine and methionine increased absolutely with 26.5, 41.4, and 13.2%, respectively.

After the ban, protein from meat and bone meal has to be replaced by plant protein sources. Differences in the content of both crude protein and essential amino acids as well as in amino acid digestibility need to be considered. Table 10 shows average analytical data from both meat and bone meal and common plant proteins.

**Table 10** Nutrients (g/kg) and AME (MJ/kg) in selected raw materials for poultry diets (DLG, 1991; Degussa, 1996)

| | DM | Crude ash | Crude protein | Lys | Met | Met + Cys | Thr | Trp | AME |
|---|---|---|---|---|---|---|---|---|---|
| Animal meal (AM) | 950 | 264 | 543 | 29.5 | 7.9 | 14.7 | 19.9 | 4.2 | 11.23 |
| Bone meal (BM) | 940 | 433 | 404 | 20.4 | 5.5 | 9.6 | 13.0 | 2.1 | 8.56 |
| AM/BM 1:1 | 945 | 348 | 474 | 25.0 | 6.7 | 12.2 | 16.5 | 3.2 | 9.90 |
| AM/BM 0.34/0.66 | 943 | 376 | 451 | 23.5 | 6.3 | 11.3 | 15.3 | 2.8 | 9.47 |
| Beans | 880 | 34 | 263 | 16.5 | 2.1 | 5.3 | 9.5 | 2.3 | 12.66 |
| Peas | 880 | 33 | 228 | 16.3 | 2.3 | 5.6 | 8.6 | 2.1 | 13.63 |
| Lupines, yellow | 880 | 45 | 386 | 17.7 | 3.0 | 8.8 | 13.7 | 2.9 | 12.88 |
| Rapeseed meal | 890 | 70 | 361 | 19.2 | 7.3 | 17.1 | 16.1 | 4.9 | 9.90 |
| Soybean meal | 880 | 59 | 451 | 28.2 | 6.5 | 13.3 | 18.0 | 5.9 | 13.04 |

On a mass basis Rodehutscord et al. (2002) stated that only soybean meal has the potential to replace protein from meat and bone meal. A mix of bone meal and animal meal (1:2) is almost equivalent to soybean meal in crude protein content. One kg of a 1:1 mix of animal meal and bone meal needs 1.05 kg soybean meal or 2.08 kg peas for protein-equivalent replacement. A replacement based on crude protein equivalents would reduce the content of crude ash and improve ME content in the complete diet in most cases. With soybean meal as the alternative, the supply of the first-limiting amino acids lysine, methionine + cystine, threonine and tryptophan to the growing animal would be improved. Similarly, protein from rapeseed meal extracted would supply comparable amounts of lysine and higher amounts of other amino acids. In this case, however, a compensation for the lower ME content is necessary. With protein-equivalent inclusion of legume seeds, the deficit in ileal digestible S-containing amino acids needs to be accounted for. For several reasons, the inclusion level for plant protein sources in compound feed is restricted. Upper limits for inclusion have been indentified for several ingredients and specific for each species (Jeroch et al., 1999). Apart from protein and amino acid content, these restrictions must be considered when protein from meat and bone meal is to be replaced, particularly in the case of rapeseed meal for laying hens.

*4.2.2 Phosphorus*

Similar as for the amino acids, alternative P sources have to be considered to ensure that the animal's P requirement is met. Sufficient P supply to growing poultry, particularly in the first half of the fattening period of broilers is difficult with feedstuffs of plant origin alone without the use of enzymes. The ban on meat and bone meal makes large amounts of P irreversibly disappearing from the food chain (Rodehutscord et al., 2002). A survey of the literature indicates that P contents vary between 16 and 42 g/kg in fish meal, between 25 to 56 g/kg in meat and bone meal, and between 17 to 35 g/kg in poultry by-products meal (Hua et al., 2005). Feedstuffs of plant origin that are rich in protein have a much lower concentration of P than animal meal or bone meal. Furthermore, the digestibility of P is lower in feedstuffs of plant origin. Meat and bone meal contributed to 57% of the supplementation of P that was needed for pigs and poultry. Table 11 shows the digestible P content of some vegetable feedstuffs without and with supplementation of microbial phytase. Supplementary microbial phytase increases digestibility of P from vegetable ingredients. Less inorganic phosphates are needed when diets are supplemented with microbial phytase.

**Table 11** Average concentration of total P and digestible P (g/kg) in feedstuffs from animal or vegetable origin (Rodehutscord et al., 2002)

| | Total-P | Digestible P | |
|---|---|---|---|
| | | Without supplemented Phytase | With supplemented Phytase |
| Animal meal | 31.0 | 25 | 25 |
| Bone meal | 61.0 | 50 | 50 |
| Soybean meal, solvent extracted | 6.5 | 2.3 | 4.2 |
| Rapeseed meal, solvent extracted | 10.5 | 3.2 | 6.8 |
| Peas | 4.2 | 1.9 | 2.7 |
| Field beans | 4.2 | 1.5 | 2.7 |
| Lupins | 4.5 | 2.2 | 2.9 |

# 5 Development of performance, behaviour and health status of laying hens in practice after the ban

## 5.1 Performance

Plant proteins are generally nutritionally imbalanced. Unless supplemented with animal proteins or free amino acids, plant-based diets may not meet the requirements of certain critical amino acids and vitamin $B_{12}$ for the production of eggs and meat (Ravindran and Blair, 1993). Animal protein supplements are decidedly superior to oilseed meals as sources of essential amino acids, particularly of lysine, the first limiting amino acid in cereal-based diets. For this reason animal proteins were normally used to balance the amino acid contents of diets rather than as major sources of protein. Bozkurt et al. (2004) conducted an experiment to study the effect of inclusion of meat and bone meal (containing a high ash and low crude protein content to conventional corn-soy based diets) on performance of laying hens at old age. In the experiment forced moulted 84-week old laying hens were used. Meat and bone meal was included at 2, 4 or 6% to a corn-soybean meal based diet. The experimental diets were formulated to be isoenergetic and isonitrogenous and also equal in calcium and phosphorus levels. Significant effects of dietary treatments on egg production and egg weight were observed (Table 12). The inclusion of 2.0% meat and bone meal in the diet increased ($P < 0.05$) hen-day egg production above that of the control diet. However, the inclusion of meat and bone meal in excess of 2.0% had no additional beneficial effect on egg production. Egg weight was reduced significantly with the inclusion of meat and bone meal at 2.0, 4.0 and 6.0% to the diet, compared to the control diet. The average egg weight of the control group was 1.0 g heavier than that of the hens receiving the diets containing meat and bone meal. On the other hand, there were no differences ($P > 0.05$) in egg mass, feed intake and feed conversion ratio between the treatments over the 20 weeks period.

**Table 12** The effect of meat and bone meal (MBM) inclusion to the diet on egg production (%), feed consumption (g), feed conversion ratio (g feed/g egg), egg weight (g) and egg mass (g) (Bozkurt et al., 2004)

| Parameters | Control | MBM levels, % | | | Pooled | Probability |
|---|---|---|---|---|---|---|
| | | 2 | 4 | 6 | SEM | |
| Egg production, hen/d, % | 63.8 [b] | 65.3 [a] | 64.3 [ab] | 64.4 [ab] | 0.37 | 0.05 |
| Egg weight, g | 70.1 [a] | 69.0 [b] | 69.0 [b] | 69.1 [b] | 0.15 | 0.00 |
| Egg mass, g/hen/d | 44.8 | 45.1 | 44.4 | 44.5 | 0.44 | 0.56 |
| Feed intake, g/hen/d | 117.4 | 117.7 | 117.5 | 116.4 | 0.54 | 0.20 |
| Feed conversion ratio, g/g | 2.6 | 2.6 | 2.6 | 2.6 | 0.03 | 0.18 |

a-b Means within rows with different superscripts differ at $P < 0.05$

Damron et al. (2001) reported, in agreement with the above mentioned results, that the inclusion of meat and bone meal up to a level of 6.0% had no negative effect on laying hen performance. In that study, rendered layer mortality was used as a feed ingredient in layer diets. Feed intake, egg production, feed conversion ratio and body weight changes of hens receiving the meat and bone meal-containing diets were superior to those on the control diet. However, a reduced egg weight was recorded when the hens received 2.5% or more meat and bone meal in their diets compared to the control diet. The adverse effect of meat and bone meal on egg weight might be associated with the amino acid concentration of meat and bone meal used in the experiments. It seems likely that the meat and bone meal used, which contained a high level of ash and a low level of crude protein, might have contained lower amino acid concentrations than those used in other studies (Parsons et al., 1997; Wang and Parsons, 1998a; Shirley and Parsons, 2001). Furthermore, the increased egg production rate of the hens receiving the meat and bone meal-supplemented diets could have had a depressive effect on egg weight (Bozkurt et al., 2004).

**Table 13** Effect of the inclusion of meat and bone meal (MBM) to a layer diet on egg quality (Bozkurt et al., 2004)

| Parameters | Control | | MBM levels, % | | | | | | Pooled | Probability |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | 2 | | 4 | | 6 | | SEM | |
| Egg specific gravity, $g/cm^3$ | 1.088 | b | 1.093 | a | 1.091 | a | 1.089 | ab | 0.002 | 0.0506 |
| Eggshell weight, g | 8.81 | | 8.94 | | 8.99 | | 9.09 | | 0.10 | 0.2521 |
| Eggshell weight ratio, % | 12.33 | b | 12.76 | a | 12.72 | a | 12.84 | a | 0.13 | 0.0330 |
| Eggshell thickness, µm | 350.68 | | 359.07 | | 359.77 | | 357.16 | | 3.30 | 0.1956 |
| Egg yolk height, mm | 19.29 | | 19.55 | | 19.68 | | 19.50 | | 0.10 | 0.0765 |
| Egg yolk colour score[1] | 12.35 | | 12.33 | | 12.51 | | 12.50 | | 0.09 | 0.3368 |
| Haugh Unit (HU) | 76.62 | b | 78.37 | b | 78.86 | b | 81.49 | a | 0.92 | 0.0028 |
| Eggshell strength, $kg/cm^2$ | 2.13 | | 2.26 | | 2.23 | | 2.32 | | 0.07 | 0.3775 |
| Cracked/broken egg ratio, % | 5.98 | a | 3.75 | b | 4.00 | b | 3.52 | b | 0.25 | 0.0001 |

[a-b] Means within rows with different superscripts differ at $P < 0.05$

[1] Roche yolk colour score: 1, light yellow; 15, orange

The specific gravity of the eggs from hens receiving the diets containing 2.0 or 4.0% meat and bone meal was higher ($P < 0.05$) than that of control hens (Bozkurt et al., 2004). Also the HU value of the eggs from the hens fed the diet containing 6.0% meat and bone meal was significantly better than that of other dietary treatments (Table 13). Damron et al. (2001) reported similar responses. However, the inclusion of meat and bone meal in the layer diet did not affect yolk colour score, eggshell thickness, eggshell weight and eggshell strength, though the percentage eggshell weight was increased ($P < 0.05$) and the ratio of broken/cracked eggs was decreased ($P < 0.01$) with the addition of meat and bone meal to the diet. Reduced eggshell quality in layers of old age is frequently observed and often associated with the corresponding increase in egg size. The results demonstrate that the inclusion of meat and bone meal to the layer diets was effective in improving eggshell quality. Since the diets were isoenergetic, isonitrogenous and contained equal levels of calcium and phosphorus, the quality of the eggshells of hens receiving the meat and bone meal containing diets was better than that of hens receiving the control diet. Waldroup and Adams (1994) and Sell and Jeffrey (1996) reported that the relative availability of phosphorus from meat and bone meal was equal to that from dicalcium-phosphate. Although the different experimental diets contained similar phosphorus and calcium concentrations, eggshell quality was improved by the inclusion of meat and bone meal. A contributing factor might be that the meat and bone meal in this study contained a much higher content of calcium and phosphorus than meat and bone meal samples analysed in industrial surveys (Johnson and Parsons, 1997; Parsons et al., 1997; Wang and Parsons, 1998b; Wang and Parsons, 1998c). Body weight and liveability of the hens from 84 wk to 104 wk of age were not affected ($P > 0.05$) by the inclusion of meat and bone meal to the layer diet.

### 5.2 Behaviour and health status

In recent years, the poultry industry has become increasingly dependent on plant protein sources, in particular on soybean meal, and it has been suggested that this trend (and/or the concomitant absence of animal protein in layer diets) might be causally related to increased feather pecking and cannibalism. In the "Report on the Welfare of Laying Hens", the Farm Animal Welfare Council (1997) stated (without any conclusive evidence) that 'lack of animal protein in the diet predisposes the flock to injurious pecking leading to cannibalism and death'. They recommended further research to identify and quantify factors in animal protein responsible for reducing injurious pecking behaviour in laying hens. So far, an effect of dietary protein source on the incidence of pecking damage in layers has never been demonstrated experimentally, although there have been anecdotal reports of outbreaks of feather pecking and cannibalism after changes in the diet from mainly animal to mainly plant protein (McKeegan et al., 2001). However, McKeegan et al. (2001) have used fish meal as an animal protein source.
To prevent feather pecking behaviour, feed producers often add some animal protein (e.g. fish meal, meat and bone meal or milk protein sources) to the diet (Hadorn et al., 1998). It has been suggested that any suppressive effect on feather pecking induced by animal protein is due to something beneficial found only in these protein sources, for instance vitamin $B_{12}$ (McKeegan et al., 2001). However, it is also conceivable that a detrimental compound in plant protein sources could increase feather pecking behaviour. As an example, phytoestrogens could elevate plasma oestradiol concentrations and affect bird behaviour (McKeegan et al., 2001). Since the ban on meat and bone meal in Europe, the diets of laying hens contain mainly vegetable proteins. In practice, farmers expect a higher occurrence of cannibalism as a result of using vegetable diets; some examples were given by Curtis and Marsh (1992). Diets based on fish meal or soybean meal protein were fed to layer pullets up to 24

weeks of age (McKeegan et al., 2001). Greater numbers of vigorous pecks/pulls were observed in the plant protein groups throughout the experiment, although they were only significantly higher from week 13 to 16. Pecking damage scores, plasma oestradiol and progesterone, and egg production, however, were unaffected by diet. Laying hens that were fed diets with exclusively vegetable protein sources, such as extracted soybean meal, peas, faba beans and extracted sunflower seed tended to a higher mortality rate due to feather pecking compared with laying hens fed a diet with 4% meat and bone meal (Richter and Hartung, 2003). Laying hens fed diets with 4% meat and bone meal had a better plumage condition at 56 weeks of age than laying hens fed diets with only vegetable protein sources or a diet with three sources of animal protein (3% meat and bone meal, 3% blood meal and 1.5% fish meal) (Pfirter and Walser, 1998). Performance and mortality (including cannibalism) were unaffected by feeding diets with either animal (herring and meat meal) or vegetable (soybean meal extracted) protein sources (Hadorn et al., 1998; Hadorn et al., 1999). Also feeding diets based on either vegetal (soybean meal), animal (blood meal, fish meal and hydrolysed feather meal) or semi-purified (casein) protein to growing bantams did not result in differences in pecking damage scores between treatments (Savory, 1998; Savory et al., 1999). The effect of different protein sources on feather pecking behaviour is summarised in Table 14. Although practical evidence suggests a higher incidence of feather pecking in laying hens fed vegetable protein diets, no confirmation of this hypothesis can be found in literature.
Higher inclusion rates of vegetable protein sources increase the proportion of fermentative degradable carbohydrates (oligosaccharides) in the diet which may result in a high fermentative activity in the gut and herewith adversely affecting gut health.

**Table 14** Effect of protein source (animal versus vegetable protein) on plumage condition, occurrence of feather pecking and mortality in birds

| Type of bird | Period of age (weeks) | Beak-trimmed | Protein source | Plumage Condition [1] | Level of feather pecking | Mortality (%) | Authors |
|---|---|---|---|---|---|---|---|
| Laying hens: | 21 – 72 | Unknown | Soybean meal | 9.2 | Not recorded | 3.3 | Hadorn et al., 1998; |
| LSL White | | | Fish meal/meat meal | 9.2 | | 2.2 | Hadorn et al., 1999 |
| Bantams | 0 – 6 | Unknown | Soybean meal (30%) | 6.1 | Not recorded | Not recorded | Savory et al., 1999 |
| | | | Fish meal (6%), Blood meal (3.2%), Feather meal (5.2%) | 4.7 | | | |
| | | | Casein (8.4%) | 5.7 | | | |
| Laying hens: | Period of | Unknown | Meat and bone meal (4%) | 4.1 | Not recorded | Not recorded | Pfirter and Walser, 1998 |
| ISA brown, | 40 weeks | | Vegetable protein sources | 3.7 | | | |
| Lohmann brown | | | Meat and bone meal (3%), blood meal (3%), fish meal (1.5%) | 3.2 | | | |

[1] Original data recalculated to a scale of 0 to 10 where 0 indicates almost naked and 10 an intact plumage.

# 6 Mode of action of processed animal protein on performance and health

## 6.1 Biological activity

Biologically active peptides have been identified in many food resources, of both vegetable and animal origin. Peptides are short amino acid segments that have biological activity, providing that they are present at the absorptive site of the gut both "intact" and "active". Limited information is available on the presence of bioactive peptides in meat and bone meal. Biologically active peptides are specific protein fragments that have influence on metabolic processes and ultimately may have a positive effect on health (Ovelgonne et al., 2007). Bioactive peptides have specific bio-functions after they are released from the parent protein source, either by digestion or prior to consumption by food processing. Once liberated, they are capable of affecting a range of physiological and metabolic processes, such as immune response, behaviour, hormonal and neurological response, and gastrointestinal function. Bioactive peptides are normally comprised of 3 – 20 amino acids residues (Clare et al., 2003). Some peptides that have been isolated and identified have specific potential, such as: antimicrobial and immunomodulatory function, Angiotensin Converting Enzyme (ACE) inhibition (anti hypertension), antioxidant activity and opioid peptides. Some of the peptides that have been identified are glycomacropeptides, immunoglobulins, lactoferrins and lysozome. The discovery of carnosine as well as ACE inhibitory peptides in meat and fish products has led to the hypothesis that meat and bone meal may have the potential to be a significant source of bioactive peptides (Ovelgonne et al., 2007). Improved nutrient retention from meat and bone meal will reduce the levels of undigested proteins reaching the lower intestine and causing pathogen proliferation (i.e. reducing the incidence of necrotic enteritis). The role of meat and bone meal as a source of bioactive peptides needs to be studied further.

## 6.2 B Vitamins

Meat meal supplies important B vitamins, particularly thiamine (vitamin $B_1$), as a supplement to rations . Vitamin $B_1$ is important as co-enzyme in fat and carbohydrate metabolism.

In literature it has been assumed that any suppressive effects on pecking induced by fishmeal are due to something beneficial found only in animal protein sources, such as the 'animal protein factor' vitamin $B_{12}$ (Bolton and Blair, 1974).

## 6.3 Phytoestrogens in plant protein sources

Phytoestrogens, plant compounds which mimic steroidal oestrogens, are found in many plant species, some of which are used as feedstuffs for poultry (Knight and Eden, 1995; Sheehan, 1995). Soybeans in particular are a rich source of the isoflavonoid phytoestrogens genistein and diadzein, which have oestrogenic potencies of 0·084% and 0·013% compared to oestradiol. Despite these relatively low potencies, phytoestrogens may be present in high concentrations in some feedstuffs (1800 mg/kg isoflavones in soybeans) and could therefore be important biological compounds (Bingham et al., 1998). The biological effects of phytoestrogens appear to depend on both species and concentration. There is evidence that several phytoestrogens are biologically active in birds, such as genistein, diadzein, equol, coumestrol, and zearalenone. Adverse effects of phytoestrogens on fowl reproductive development have been reported, with laying hens fed diets containing high levels of coumestrol (an isoflavone) exhibiting late sexual maturation, depressed egg production and low egg weight (Mohsin and Pal, 1977). Importantly, elevated plasma oestradiol concentrations have been reported in laying hens fed diets containing soyabean meal (compared to a diet containing fishmeal as the main protein source) (Maurice et al., 1979; Akiba et al., 1982). The increased plasma oestradiol concentration seen in these studies suggest that phytoestrogens were acting antagonistically, increasing gonadotrophin secretion and gonadal activity in a similar way to the anti-oestrogen tamoxifen. Several authors have observed that pecking damage begins or increases at sexual maturation (Blokhuis, 1991; McKeegan, 1998; Bilcik and Keeling, 1999), when plasma concentrations of gonadal hormones are elevated. Such associations provide evidence for a relationship between hormonal state and the incidence of pecking damage.

# 7 Conclusions

The most important conclusions of this study are:
- Processed animal proteins are rich in protein (lysine and methionine), calcium and phosphorus (supplied by the bone fraction), and vitamin $B_{12}$.
- Digestibility of amino acids and availability of phosphorus is higher in animal protein sources compared with vegetable protein sources.
- The ratio of bone to soft tissue determines the ash content of processed animal proteins. A higher ratio of bone to soft tissue increases the ash content and has a negative effect on protein and energy concentrations.
- The bone fraction contains collagen which is deficient in most essential amino acids (tryptophan, sulfur amino acids and isoleucine). An increase in bone content of the raw materials negatively affects protein quality due to the high collagen content and poor amino acid balance.
- The quality of animal protein sources (availability of amino acids) is affected by heat treatment and raw material composition.
- Increasing pressure during processing of meat and bone meal decreases amino acid digestibility in laying hens.
- After the ban on meat and bone meal, the amount of soybean meal, wheat and corn was increased significantly in diets without animal products. The use of phytase was also increased to make phytic acid in vegetable feedstuffs available. Monocalcium-phosphate was included in vegetable diets to increase the calcium and phosphorus content in order to meet the requirements of the laying hen.
- Higher inclusion rates of vegetable protein sources increases the proportion of fermentative degradable carbohydrates (oligosaccharides) in the diet which may result in a high fermentative activity in the gut and herewith adversely affecting gut health.
- In research it was shown that vegetable diets compared to animal protein-enriched diets resulted in a decreased egg production rate and an increased egg weight. Egg mass was not affected. Specific gravity of the eggs, Haugh Unit, percentage eggshell weight was decreased and the ratio of broken/cracked eggs was increased in laying hens fed diets without meat and bone meal.
- An effect of dietary protein source on the incidence of pecking damage in layers has not been demonstrated experimentally, however practical evidence suggests a higher incidence of feather pecking and chronic gastroenteritis at different ages after the ban on animal protein. More research in this field should be developed.
- It is suggested that any suppressive effect on feather pecking induced by animal protein is due to beneficial compounds only observed in these animal protein sources, for instance (the animal protein factor) vitamin $B_{12}$, or bioactive peptides. However, it is also conceivable that a detrimental compound in plant protein sources could increase feather pecking behavior, for instance phytoestrogens. More research in this field should be developed.

In most papers meat and bone meal is discussed and in most cases the origin of the meat and bone meal is not described. From literature it is known that the variability of meat and bone meal and meat meal is large. Maybe meat meal from pigs may be used in poultry diets once the ban on meat and bone meal is abolished. Information on the feeding value and possible health stimulating effects of meat meal from pigs is lacking. So more research on the feeding value, behaviour and gastro intestinal health of feeding pig meat meal from pigs to laying hens should be considered. In such research it is important to describe the origin of the meat meal in detail, conditions during processing, and the chemical composition of this feedstuff should be determined prior to diet formulation.

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