The Chemical Composition and Nutritive Value of Mulberry Leaf as a Protein Source in Poultry Diets

R.A. Al-kirshi[1], A.R. Alimon[1,2], I. Zulkifli[1], M. W. Zahari[4], and A.Q. Sazili[1,3]
[1]Department of Animal Science,
[2]Institute of Tropical Agriculture,
[3] Institute of Halal Products Research, Universiti Putra Malaysia, Selangor, Malaysia
[4]Division of Livestock Production, MARDI. Serdang, Selangor, Malaysia
email:riyadh_12@yahoo.com

ABSTRACT

A study was conducted to determine the chemical composition of mulberry (*Morus alba*) leaf meal (MLM) and its nutritive value as a feed ingredient in diets of broiler and layer chickens. The crude dry matter, protein, ash, fat, crude fibre, NDF, ADF, Ca, P and gross energy contents were 89.3%, 29.8%, 11.8%, 11.1%, 32.3%, 22.8%, 0.28% 2.7% and 4220kcal/kg respectively. The amino acids composition of MLM indicates it is a good source of essential amino acids especially lysine 1.88% and leucine 2.55%. The overall ileal true amino acids availability was similar between layers and broilers. The digestibility coefficients of individual amino acids for aspartic acid, glutamic acid, tyrosine, histidine, argnine, glycine and tryptophan was significantly ($p<0.05$) higher for layers compared to those for broilers. However, the digestibility of methionine was higher ($P<0.05$) in broilers than layers.

Key words: mulberry, leaf meal, digestibility, amino acids, layers, broilers

INTRODUCTION

Mulberry grows well in the tropics and subtropics, and is reported to have excellent nutritional value. It is grown extensively for leaves, which are used for raising silkworms in the sericulture industry. Mulberry leaves are very rich in protein (15-35%), minerals (2.42-4.71% Ca, 0.23-0.97% P, 1130-2240 kcal/kg metabolitezable energy and absence of anti-nutritional factors (Omar *et al*., 1999; Sanchez, 2002, 2000; Saddul *et al*., 2003; Sarita *et al*., 2006). Its protein quality is comparable to those of soybean meal (Machii, 1989). Excellent results have been obtained with mulberry leaves as ruminant feed (Rojas & Benavides, 1994; Oviedo *et al*., 1994; Esquivel *et al*., 1996; Gonzalez, 1996). Published values on the digestibility of amino acids and chemical components for chickens are lacking. Thus the objectives of this study were to determine the nutrient composition and amino acid availability to chicks.

MATERIALS AND METHODS

Birds and Housing

Fifteen layer (Isa-brown strain) and fifteen male broiler (cobb strain) 17 weeks and 42 days old respectively were used in the experiment to determine amino acids availability of MLM. The birds were assigned to individual cages, during which time they were fed on a commercial layer and broiler finisher diet.

Diets

A basal diet (adaptation diet) which contained MLM as the sole source of dietary protein and DL- methionine was added at 0.12% in the basal diet. This diet was formulated to contain 16 % CP, 2850 kcal ME/kg. Assay diet was the same with basal diet except for the elimination of synthetic methionine. A protein-free diet was also formulated specifically to allow the determination of endogenous flows of amino acids. All diets were fed in mash form. Chromic oxide was included in all diets as an indigestible marker. The composition of the basal, assay and protein-free diet and its amino acid contents are shown in Table 1 and Table 2 respectively.

Feeding Trial

The chickens were offered the basal diet *ad libitum* for three days adaptation period, which is considered sufficient to eliminate the carry-over effect between the two different diets (Kadim *et* *al*., 2002). Following diet adaptation period, the birds were fasted for 24 hr. The birds were then allowed to consume the respective diets for a one hour period (Kadim and Moughan, 1997). Four hours after initial meal, the birds were killed by an intravenous injection of 1ml sodium pentobarbitonein the wing vein. The use of pentobarbiotone is to minimize peristaltic movements and mucosal shedding of the gastrointestinal tract, which may occur with other methods of killing. When the birds were completely immobilized, the body cavity was opened, the ileum removed from the location of Meckel's diverticulum to a point 5 cm proximal to the ileo-caecal junction and digesta gently flushed by using a syringe into a plastic container. Digesta samples were immediately stored at -20°C. The samples were subsequently freeze-dried, finely ground and stored at -20°C for further chemical analysis.

Table 1. Composition of the diets used in AA determination

Ingredient	Basal Diet	Assay Diet	Protein-free Diet
Mulberry	54.00	54.12	-
Dextrose	37.38	37.38	94.50
Corn oil	5.00	5.00	-
Salt	0.40	0.40	0.40
Dicalcium phosphate	1.50	1.50	2.50
Limestone	0.50	0.50	1.50
Choline choloride	0.30	0.30	0.30
Mineral-vitamine premix	0.50	0.50	0.50
DL-Methionine	0.12	0	0
Chromic oxide	0.30	0.30	0.30
Calculated analysis			
ME (kcal/kg)	2850	2850	3460
Crude protein%	16	16.1	0
Crude fibre %	6	6.1	0
Calcium %	2.00	2.00	1.17
Phosphorus %	0.40	0.40	0.45

*Supplied per kg diet: Fe, 35mg; Mn, 70mg; Cu, 8mg; Zn, 70mg; I, 1mg; Se, 0.25mg ; Co, 0.2mg; calcium-D-pantothenate, 8mg; folic acid, 0.5mg; D-biotin, 0.045 mg; vitamin C, 50mg; vitamin A, 8000 IU; vitamin D, 1000 IU; vitamin E, 30 IU; vitamin K3, 2.5mg; vitamin B1, 2mg; ; vitamin B2, 5mg; vitamin B6, 2mg; vitamin B12, 0.01 mg; and niacin, 30mg.

Chemical Analysis

Amino acid concentrations for ileal digesta were determined by HPLC as described by Cohen and Strydon (1994) following pre-column derivatisation with AQC reagent (6-aminoquinolyl-N-hydroxy-succinimdyl carbamate, waters, USA). Cystine and methionine were analysed as cystic acid and methionine sulfone by oxidation with performic acid for 16h at 0ºC and neutralization with hydrobromic acid before hydrolysis. Tryptophan contents were determined following alkaline hydrolysis of sample with 4.3N. $LiOH.H_2O$ for 16h at 120°C and neutralization with 6N HCl. Rest of the AA were hydrolyzed by 5ml 6N HCl for 22 hours at 110°C. Chromium was determined according to Saha and Gilbreath (1991).

Table 2. Dry matter, crude protein and amino acid contents (%) in diets and mulberry leaf meal used in metabolic study

	Basal diet	Assay diet	MLM
Dry matter	93.11	93.11	89.30
Crude protein	16.00	16.10	29.80
Aspartic acid	1.50	1.50	3.06
Glutamic acid	1.70	1.70	3.33
Serine	0.70	0.70	1.22
Glycine	0.80	0.80	1.57
Histidine	0.44	0.44	0.69
Arginine	1.30	1.30	1.80
Threonine	0.79	0.79	1.31
Alanine	0.70	0.70	1.54
Proline	0.60	0.60	1.30
Tyrosine	0.30	0.30	0.82
Valine	0.96	0.96	1.76
Methionine	0.38	0.25	0.52
Cyctine	0.15	0.15	0.30
Isoleucine	1.10	1.10	1.43
Leucine	1.80	1.80	2.58
Phenylalanine	1.20	1.20	1.94
Lysine	1.10	1.10	1.88
Tryptophan	0.15	0.15	0.27

Calculations

Apparent digestibility of the assay diet was calculated using the following equation (Kadim and Moughan, 1997):

Apparent AA digestibility (AID) (%) =

$$\text{AA concentration in diet} - \frac{\text{AA output in ileum}}{\text{AA concentration in diet}} \text{x}100\%$$

True AA digestibility of the assay diet was calculated using the following equation (Kadim and Moughan, 1997):

True AA digestibility (%) =

$$\frac{\text{AID+endogenous AA Output}}{\text{AA concentration ion in diet}} \text{ x } 100\%$$

Where: AID: Apparent amino acid digestibility

Statistical Analysis

Data were subjected to analysis of variance (ANOVA) using SPSS (2005). Duncan's multiple range test was used to compare the means.

RESULTS AND DISCUSSION

The chemical and amino acid composition of the MLM are presented in Table 3. The amino acids content of mulberry leaves was comparable to those reported by Machii (1989) and Coto (1996).

Table 3. Chemical composition of mulberry leaves (%DM)

Nutrients	Content
Dry matter, %	89.30
Crude protein, %	29.80
Ether extract, %	5.57
Crude fiber	11.10
Gross energy, kcal/kg	4220
Ash, %	11.8
Neutral detergent fiber, %	35.80
Acid detergent fiber, %	28.00
Hemicellulose, %*	7.80
Calcium, %	2.73
Phosphorus, %	0.28
Amino acids	
Aspartic acid	3.06
Glutamic acid	3.33
Serine	1.22
Glycine	1.57
Histidine	0.69
Arginine	1.80
Threonine	1.36
Alanine	1.54
Proline	1.31
Tyrosine	0.82
Valine	1.77
Methionine	0.52
Cyctine	0.30
Isoleucine	1.43
Leucine	2.58
Phenylalanine	1.94
Lysine	1.88
Tryptophan	0.27

* Neutral detergent fibre-acid detergent fibre

Table 4 shows a comparison of the composition of amino acids from mulberry leaves with the amino acid requirements of chicks as given by the NRC (1994). It was found that mulberry leaves have a good essential amino acid profile relative to meet the chicks requirements. However, cystine is deficient in mulberry leaves.

Table 4. Composition of amino acids from mulberry leaf meal (DM basis) in relation to the requirements for growth of chicks

Amino acid	N.R.C. requirement (%)	Mulberry leaf meal (%)
Argnine	1.44	1.80
Lysine	1.20	1.80
Histidine	0.35	0.69
Phenylalanine	0.72	1.94
Tyrosine	0.62	0.82
Methionine	0.50	0.52
Leucine	1.35	2.58
Isolecine	0.80	1.43
Valine	0.82	1.76
Threonine	0.80	1.31
Tryptophan	0.23	0.27
Cystine	0.43	0.30

True AA digestibility of mulberry leaf meal is shown in Table 5. The amino acid digestibility of MLM ranged from 65.54% for lysine in broilers to 81.99% for tryptophan in layers.

Table 5. True amino acid digestibility (%) of mulberry leaf meal of layer and broiler chickens

Amino acids	Layers	Broilers	S.E.M
Aspartic acid	75.85[a]	71.69[b]	0.37
Glutamic acid	75.95[a]	73.78[b]	0.38
Serine	77.60	80.85	0.40
Glycine	78.30[a]	72.71[b]	0.35
Histidine	68.80[a]	66.55[b]	0.67
Arginine	79.15[a]	75.33[b]	0.55
Threonine	72.73	73.16	0.45
Alanine	73.21	72.99	0.56
Proline	73.79	73.25	0.20
Tyrosine	75.83[a]	69.47[b]	0.79
Valine	73.82	74.03	0.41
Methionine	76.21[b]	79.72[a]	0.85
Cyctine	74.67	74.36	1.14
Isoleucine	79.38	79.72	0.37
Leucine	76.62	77.02	0.71
Phenylalanine	73.75	75.18	0.86
Lysine	66.74	65.54	0.83
Tryptophan	81.99[a]	79.10[b]	0.92
Over all mean	74.23	74.53	0.69

Overall amino acids digestibility for layers and broilers fed MLM was 74.23% and 74.53% respectively. There was no significant difference in overall amino acids digestibility between the two classes of birds. However layers fed MLM was significantly ($p<0.05$) digested more aspartic acid, glutamic acid, tyrosine, histidine, argnine, glycine and tryptophan. The digestibility of methionine is significantly higher ($p<0.05$) in broiler than layer. The class of bird influenced the digestibility coefficients of amino acids in mulberry leaf meal. However, the average overall digestibility of amino acids was similar between layers and broilers. Effects on the digestibility of individual amino acids were higher in layers. The digestibility of glycine, histidine, arginine, aspartic, glutamic, tyrosine and tryptophan was higher in layers compare to broilers, while that of methionine, was higher in broilers. The digestibility of other amino acids was similar between the two classes of birds. The poor individual amino acid digestibility in broilers feeding mulberry leaves attributed to the high level of NDF in mulberry leaves. This is in agreement with findings that the nutrients in diets containing high fibre levels are poorly digested in broiler (Annison *et al*., 1997).

CONCLUSIONS

Mulberry (*Morus alba*) leaf meal can be used as a potential source of protein in poultry production. The true ideal digestibility of amino acids in MLM is influenced by the class of birds. This need further research in order to confirm this finding.

ACKNOWLEDGMENTS

The authors thank the staff of the Poultry Unit and Nutrition Laboratory of the Department of Animal Science, University Putra Malaysia for their technical assistance.

REFERENCES

Annison, G. Mughan, P.J. Hendriks, W.H. and Bourne, S. 1997. The non-starch polysaccharides of wheat middling by-products. Proc. Aust. Poultry Science Symp 9: 244.

Cohen, S.A. and Strydon, D.J. 1994. Comparision of amino acid analysis by Phenylisothiocyanate and 6-Aminoquinoly-N-hydroxysuccinidyl carbamate precolumn derevitisation. Analytical Chemeistry. 222: 19-28

Coto, O.M. 1996. Importancia nutritiva de la Morera (*Morus alba*). San Andrés, El Salvador, Centro Nacional de Tecnología Agropecuaria y Forestal. Ministerio de Agricultura y Ganadería. 6 pp. (mimeo).

Esquivel, J., J.E., Benavides, I. Hhernandes, J. Vasconcelos, J. Gonzallez and E. Espinoza. 1996. Effecto de la sustitucion de concentrado con morera (*Morus alba*) sobre la produccion de leche de vacas en pastoreo. In: Resumenese. Taller Internacional "Los arboles en la produccion ganadera" EEPF "Indio Hatuey", Matanazas, Cuba, P:25.

Gonzalez, J. 1996. Evaluacion de la calidad nutricional de la morera (*Morus sp.*) fresca y ensilada, co bovions de ongorda. Tesis mag. Sc. Turrialba, C.R. CATIE, 84p.

Kadim, I. T., and Moughan, P.J. 1997. Development of an ileal amino acid digestblity assay for the growing chicken-effect of time after feeding and site of sampling. British Poultry Science. 38:89-95.

Kadim, I. T., Moughan, P.J. and Ravindran, V. 2002. Ileal amino acid digestblity assay for the growing chicken- comparison of ileal and excreta amino acid digestblity in the chicken. British Poultry Science. 44: 588-568.

Machii, H. 1989. Varietal differences of nitrogen and amino acid contents in mulberry leaves. Acta sericologica et entomologica. Japan.pp 51-61.

Omar, S.S., Shayo, C.M. and Uden, P. 1999. Voluntary intake and digestibility of mulberry (*Morus alba*) diets by growing goats. Tropical Grasslands. 33:177-181.

Oviedo, F.J., Benavides, J.E. and Vallejo, M. 1994. Evaluacion bioeconomica de un modulo agroforestal concabras en el tropico humedo. In:Benavides, J.E. (ed.) Arbolesy arbustos forrajeros en America Central. Volumen I. CATIE, Turrialba,Costa Rica, 601-629.

Rojas, H. and Benavides, J.E. 1994. Produccion deLeche de cabras alimentadas con pastro ySuplementadas con altos niveles de morera (*Morus*Sp.) In: Benavides, J.E. (ed.) Arbolesy arbustos Forrajeros en America Central. Volumen I. CATIE, Turrialba, Costa Rica, P: 305-317.

Saddul, D., Jelan, Z. A. Liang, J. B. and Halim, R. A. 2004. The potential of *Morus alba* as a fodder crop: The effect of plant maturity on yield, persistence and nutrient composition of plant fractions. Asian-Aust. J. Anim. Sci. 17 (12): 1657-1662.

Saha, D.C. and Gilbreath, R. L. 1991. Analytical recovery of chromium from diet and facese determined by calorimetry and atomic absorption spectrophotometry. J Sci. Food Agri. 55, 433-446.

Sanchez, M.D. 2000. Mulberry: an exceptional forage available almost worldwide. World Animal Review. 93, 1–21, FAO, Rome.

Sánchez, M.D. 2002. Mulberry for animal production. FAO Animal Production and Health Paper 147. Food and Agriculture Organization of the United Nations, Rome, Italy.

Sarita, S., Rashmi, K., Anju, T. and Rajesh Prakash. 2006. Nutritional quality of leaves of some geno types of mulberry (*Morus alba*). Int. Journal of Food Sci. and Nutrition. 57(5/6): 305-313.

SPSS. 2005. Statistical Package for Social Scientists. SPSS Inc., Chicago, IL.