

ISSN 1682-8356
ansinet.org/ijps



INTERNATIONAL JOURNAL OF
POULTRY SCIENCE

ANSI*net*

308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorijps@gmail.com

Broiler Breeder Age and Dietary Energy Level on Performance and Pancreas Lipase and Trypsin Activities of 7-days Old Chicks

Alex Maiorka¹, Ana Vitoria Fischer da Silva¹, Elizabeth Santin¹,
João Martins Pizauro Jr.² and Marcos Macari²

¹Universidade Federal do Paraná - UFPR 80035-050 Curitiba PR Brazil

²Faculdade de Ciências Agrárias e Veterinárias - Unesp, 14884-900 Jaboticabal SP Brazil
E-mail: besantin@aol.com

Abstract: The objective of this study was to evaluate the effects of broiler breeder age and the dietary energy level (2,900 and 3,200 kcal ME/kg) on performance and pancreas lipase and trypsin activities of chicks during the first week of life. The birds from the older breeders had higher feed intake ($P = 0.001$) and weight gain ($P = 0.001$) than chicks from the younger broiler breeders. The pancreas enzymes activities were not affected by broiler breeder age, however it was influenced by the dietary energy level. Chicks fed diet with high energy content showed higher lipase activity than those submitted to low dietary energy level ($P = 0.013$) and trypsin activity was not affected by energy diet. No improvement on chicks performance was observed, suggesting that is not necessary to feed diets with high energy level during the first week of age.

Key words: Broiler breeder age, enzyme activity, dietary energy, lipase, trypsin

Introduction

During avian embryo development, the nutrients are supplied by egg and after hatching the birds start using the nutrients supplied by a complex diet. This change in the way to obtain nutrients must have an adaptation period in the gastrointestinal tract of birds. It is speculated that the first week of life is very important to full development of the intestinal tract, until the relative size of the intestines and the enzymes production are optimized.

Lipid is not efficiently used by young chicks (Seel, 1996). Maiorka *et al.* (1997) reported that different dietary energy levels fed during the first week of life did not affect performance of the birds until 21 days of age, despite the different energy intake.

Lipid digestion and absorption requires many physiological variables to occur such as the presence of bile salts, pancreas lipase, colipase and binding protein of fatty acids. During the early period after hatching, avian species show an immature entero-hepatic circulation and this results in negative effects on lipid digestion and absorption (Serafin and Nesheim, 1970; Jeanson and Kellogg, 1992).

On the other hand, according to Wilson (1991), there is a high correlation between egg weight and chick weight at hatching. As the broiler breeder ages, it produces bigger follicles, which results in larger eggs with larger yolks (Zakaria *et al.*, 1983). Therefore, eggs from older broiler breeders are heavier than those from younger broiler breeders. This means, that chicks from older broiler breeders have higher weights at hatching. Some studies (Applegate *et al.*, 1999; Applegate and Lilburn, 1999) also showed differences on intestinal morphology between poult embryos of broiler breeders of different ages and after hatching poult from older breeders

Table 1: Composition of experimental diets

Ingredients (%)	Low energy	High energy
Yellow corn	54.60	54.60
Soybean meal	28.99	28.99
Gluten - 60	7.50	7.50
Soybean Oil	1.17	4.58
Dicalcium phosphate	1.70	1.70
Limestone	1.43	1.43
Salt (NaCl)	0.45	0.45
Vitamin and trace mineral mix ^A	0.65	0.65
DL-Methionine	0.10	0.10
Sand	3.41	-
Calculated analysis		
Crude protein, %	22.00	22.00
ME, kcal/kg	2,900	3,200
Calcium, %	1.00	1.00
P available, %	0.50	0.50
Sodium, %	0.20	0.20
Methionine, %	0.51	0.51
Methionine + Cystine, %	0.88	0.88
Lysine, %	1.10	1.10

^AProvided per kilogram of diet: vitamin A (11,925 IU); vitamin D₃ (2,250 IU); vitamin E (9 IU); vitamin K₃ (1.8 mg); vitamin B₁₂ (0.02 mg); thiamin (1.1 mg); riboflavin (9 mg); pirodoxine (1.8 mg); biotin (0.1 mg); Pantothenic acid (9.9 mg); niacin (38.25 mg); Folic acid (0.9 mg); Choline (680 mg); Iodine (1.2 mg); Selenium (0.18 mg); Iron (70 mg); Copper (10 mg); Zinc (60 mg); Manganese (70 mg)

showing higher villi height and higher capacity to metabolize glucose than poult from younger breeders, suggesting that birds from younger breeders are not fully adapted to the metabolic changes caused by post-hatching feed.

The objective of this study was to evaluate the performance and pancreas enzymes activities of broilers

Table 2: Feed intake (g), body weight gain (g) and feed conversion (g/g) from 1 to 7 days of age

Broiler breeder age (weeks)	EM (kcal EM/kg of the diet)	Feed intake (g)	Body weight gain (g)	Feed conversion (g/g)
30	2,900	127	102	1,250
30	3,200	135	107	1,181
60	2,900	137	115	1,194
60	3,200	147	117	1,266
Principal effects				
30 weeks		131	104	1,216
60 weeks		142	116	1,230
2,900 kcal EM/kg		133	109	1,221
3,200 kcal EM/kg		141	112	1,224
Probability				
Broiler breeder age (A)		0.001	0.001	0.712
Energy (B)		0.156	0.571	0.958
A x B		0.163	0.462	0.177

Table 3: Liver, gizzard plus proventriculus, pancreas and intestines weight expressed as percentage of body weight (%) at 7 days of age

Broiler breeder age (weeks)	EM (kcal EM/kg of the diet)	Liver (%)	Gizzard + Proventriculus (%)	Pancreas (%)	Intestines (%)
30	2,900	3.52	6.41	0.435	7.76
30	3,200	3.24	6.69	0.385	7.73
60	2,900	3.08	6.43	0.434	7.36
60	3,200	3.18	6.34	0.355	7.81
Principal effects					
30 weeks		3.38	6.55	0.410	7.75
60 weeks		3.13	6.39	0.395	7.59
2,900 kcal EM/kg		3.30	6.51	0.425	7.77
3,200 kcal EM/kg		3.21	6.43	0.380	7.56
Probability					
Broiler breeder age (A)		0.091	0.286	0.445	0.460
Energy (B)		0.361	0.548	0.091	0.338
A x B		0.390	0.216	0.475	0.271

Coming from broiler breeders of different ages and submitted to different dietary energy levels during the first week of age.

Materials and Methods

A total of 240 day-old male Cobb-500 chicks, coming from eggs of 30-wk and 60-wk broiler breeders, were used. The average of star weight to chicks from 30-wk broiler breeder was 45.63 ± 2.79 and 41.80 ± 2.84 to chicks from 60-wk broiler breeders. Immediately after hatching, chicks were housed into battery cages in an environmentally controlled room where ambient temperature was maintained at thermoneutrality with continuous light.

Diets were formulated based on corn and soybean meal and their composition are shown in Table 1. Feed and water were supplied *ad libitum*. At 7 days of age, chicks and feeders were weighed to obtain feed intake, weight

gain and feed conversion. Five birds per treatment were slaughtered by cervical dislocation. Liver, gizzard + proventriculus, pancreas and small intestine were weighed ($g \pm 0.01$). Gizzard + proventriculus and intestines were opened to remove feed residues. Weights of these organs were calculated as percentage of chick body weight. Pancreas was taken and immediately frozen in liquid nitrogen and kept at -70°C .

Enzymatic assay: Pancreas was homogenized in Ultra-Turrax in 500 mM Tris-HCl buffer containing 50 mM CaCl_2 (1:20 w/v), pH 8.0, at 4°C . Homogenate was centrifuged under refrigeration (4°C) at 14,000g for 30 minutes. An aliquot of pancreatic supernatant was used for immediate lipase determination. The remaining

Table 4: Pancreas enzymes activities (lipase and trypsin) of chicken at 7 days of age

Broiler breeder age (weeks)	EM (kcal EM/kg of the diet)	Lipase*	Trypsin*
30	2,900	9.38	36.76
30	3,200	13.21	43.29
60	2,900	11.81	33.49
60	3,200	11.83	40.74
Principal effects			
30 weeks		11.31	40.03
60 weeks		11.83	37.13
2,900 kcal EM/kg		10.61	42.02
3,200 kcal EM/kg		12.53	35.12
Probability			
Broiler breeder age (A)		0.477	0.529
Energy (B)		0.013	0.141
A x B		0.145	0.938

+: $\mu\text{mol}/\text{min}/\text{mg}$ of protein (specific activity)

*: $\text{nmol}/\text{min}/\text{mg}$ of protein (specific activity)

supernatant was frozen in liquid nitrogen and stored at $-70\text{ }^{\circ}\text{C}$ until tripsis activity determination.

Pancreas lipase activity was assessed by titration (Tietz and Fiereck, 1966), using the olive oil emulsion (SIGMA[®]) substrate and the colipase excess extracted from poultry pancreas. One unit of enzymatic activity was defined and expressed as the quantity of enzyme that release one μmol of fatty acid per minute.

Activation of pancreatic trypsinogen was accomplished by pre-incubation period with 0.08 units of Enterokinase (SIGMA[®]) for 30 minutes. After the activation, the trypsin activity was determined at $37\text{ }^{\circ}\text{C}$ according to Kakade *et al.* (1974), using N- α -benzoyl-L-arginine-p-nitroanilide (L-BAPNA, SIGMA[®]) as substrate. One unit of enzyme activity was defined and expressed as the quantity of enzyme that release one μmol of p-nitroanilide/min, at $37\text{ }^{\circ}\text{C}$, since enzymes activities were expressed as specific activity ($\mu\text{mol}/\text{min}/\text{mg}$ protein) the protein concentration in pancreas tissue was determined by the procedure described by Hartree (1972), using bovine serum albumin as standard.

Experimental design and statistical analysis: A completely randomized experimental design was used, in a factorial scheme 2×2 (2 ages of broiler breeder, 30 and 60 weeks and 2 energy levels, 2,900 and 3,200 kcal ME/kg). The experimental unit consisted of a cage with 12 birds, with 5 replicates per treatment, with a total of 20 cages and for pancreas enzyme analysis one bird was one experimental unit. Data were submitted to analysis of variance using the General Linear Model (GLM) procedure of SAS.

Results and Discussion

Chicks from older broiler breeders (60 weeks of age) showed higher feed intake ($P = 0.001$) and body weight

gain ($P = 0.001$), than chicks from younger broiler breeders during the first week of life (Table 2). The relationship of egg size and chick size at hatching was reported by Shanawany (1987) and according Wilson (1991) for each additional gram in egg weight, the chick has an increment in two to 13 grams in body weight at hatching, which remains until the six weeks of life of the broiler. Leeson and Summers (2000) also reported that one-gram plus in egg weight could result in ten to 15 grams at forty days of age, being more evident in males. The relative weight of liver, pancreas, gizzard + proventriculus and intestine and length of intestine were not influenced by the factors studied until the seven days of age of broilers (Table 3). Also, no interaction was observed between broiler breeder age and dietary energy levels of broiler initial diets on any of the parameters analyzed in the present study.

Although the lipids are the most important source of energy for the embryo during the incubation period, in the present study, the dietary energy level after hatching did not influence the performance of birds during the first week of life. It is known that 80% of yolk lipids are mobilized and absorbed in the last seven days of incubation, but the capacity to digest and absorb lipids from diets after hatching is still not well developed, but it increase as the bird aged (Krogdahal, 1985). This immaturity to digest and absorb lipids could be an explanation to the findings of this study, since no significant difference was observed on performance according to the dietary energy level. Similar results were previously reported by Maiorka *et al.* (1997), irrespective of broiler breeder ages.

The activity of pancreas lipase was higher ($P = 0.013$) in birds fed diets containing high energy level (Table 4). This data agrees with Krogdahal (1985), who observed a 10-fold increment in lipase activity when to the broiler diet was added high oil concentration. Krogdahl and Sell (1989) reported that pancreas lipase activity could not be a limiting factor to fat digestion in the gut, as bile salts are. The immature entero-hepatic circulation could affect negatively the digestion of lipids, since it reduces the fat emulsification (Serafin and Nesheim, 1970). And, perhaps, this reduced physiological capacity could explain why the increase in lipase level in the present study did not result in an improvement of chick performance. Carew *et al.* (1972), also observed similar findings when vegetal oil or animal fat were used in broiler chicks diets.

According to the results of the present study and other related reports, can be suggested that the immature entero-hepatic circulation might be the most important limiting factor of the young bird to use lipids of the diet, since that lipase enzyme concentration and activity can be modulated by the increments of lipids in the gut.

Conclusions: The results showed that broiler breeder

age affected the performance of chicks at 7-days of life and that the use of high dietary energy levels due to lipid increment did not improve the chick performance. On the other hand, despite lipase activity had increased due to dietary energy level, the physiological limiting factor to fat digestion and absorption, seems to be related to bile salts secretion in the gut.

Acknowledgements

The authors thank Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP– Proc. 98/11304-9) for financial support.

References

- Applegate, T.J., J.J. Dibner, M.L. Kitchell, Z. Uni and M.S. Lilburn, 1999. Effect of turkey (*Meleagris gallopavo*) breeder hen age and egg size on poult development. 2. Intestinal villus growth, enterocyte migration and proliferation of the turkey poult. *Comp. Biochem. Physiol.*, 124B: 381-389.
- Applegate, T.J. and M.S. Lilburn, 1999. Effect of turkey (*Meleagris gallopavo*) breeder hen age and egg size on poult development. 1. Intestinal growth and glucose tolerance of the turkey poult. *Comp. Biochem. Physiol.*, 124B: 371-380.
- Carew, L.B., R.H. Machemer Jr and R.W. Sharp Jr., 1972. Fat absorption by the very young chick. *Poult. Sci.*, 51: 738-742.
- Hartree, E.F., 1972. Determination of protein. A modification of the Lowry method that gives a linear photometric response. *Analytical Biochem.*, 48: 422-7.
- Jeanson, S.E. and T.F. Kellogg, 1992. Ontogeny of taurocholate accumulation in the terminal ileal mucosal cells of young chicks. *Poult. Sci.*, 71: 367-372.
- Kakade, M.L., J.J. Rackis and J.G. Mcghee, 1974. Determination of trypsin inhibitor activity of soy products: A collaborative analysis of an improved procedure. *Cereal Chem.*, 51: 376-82.
- Krogdahal, A., 1985. Digestion and absorption of lipids in poultry. *J. Nutr.*, 115: 675-685.
- Krogdahal, A. and J. Sell, 1989. Influence of age on lipase, amylase and protease activities on pancreatic tissue and intestinal contents of young turkeys. *Poult. Sci.*, 68: 1561-1568.
- Leeson, S. and J.D. Summers, 2000. Commercial poultry nutrition. University Books, Guelph.
- Maiorka, A., J.L. Lecznieski, H.A. Bartels and A.M. Penz Jr., 1997. Efeito do nível energético da ração sobre o desempenho de frangos de corte de 1-21 dias de idade. In: *Anais Conferência Apinco'97 de Ciência e Tecnologia Avícolas*, São Paulo: Facta.18.
- Serafin, J.A. and M.C. Nesheim, 1970. Influence of dietary heat-labile factors in soybean meal upon bile acid and turnover in the chick. *J. Nutr.*, 100: 786-796.
- Seel, J.L., 1996. Physiological limitations and potential for improvement in gastrointestinal tract function of poultry. *J. Appl. Poult. Res.*, 5: 96-101.
- Shanawany, M.M., 1987. Hatching weight in relation to egg weight in domestic birds. *World's Poult. Sci. J.*, 43: 107-115.
- Tietz, M.W. and E.A. Fiereck, 1966. A specific method for serum lipase determination. *Clinica Chimica Acta*, 13: 352-358.
- Wilson, H.R., 1991. Interrelations of egg size, chick size, posthatching growth and hatchability. *World's Poult. Sci. J.*, 47: 5-20.
- Zakaria, A.H., T. Miyaki and K. Imai, 1983. The effect of aging on ovarian follicular growth in laying hens. *Poult. Sci.*, 62: 670-674.