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POTENTIAL OF MICROBIAL *PSEUDOMONAS SP.* LIPASE ON BROILER PERFORMANCE

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Key words:

Pseudomonas sp. Lipase, Fat Utilization, Broilers.

Summary:

An experiment was conducted to test the dietary effect of 5 levels of liquid form of *Pseudomonas sp*. Lipase in broiler chickens (0, 50, 100, 200 and 400 U/g of diet) in a corn-soybean based diet containing 8% tallow from 0-35 days of age. Lipase addition had a negative impact on body weight gain, feed intake, feed to gain ratio and fat utilization. Lipase activity of duodenal + jejunal contents decreased at 21 and 35 days of age.

Introduction:

The ability of young chickens and turkeys to utilize fat is not universally high. In particular, saturated fats are poorly utilized by young birds (Leeson and Atteh, 1995). The reason for the poor fat utilization has been speculated to be due to a deficiency of bile salts and/or pancreatic lipase. However, the evidence to date suggests that bile salt insufficiency is not the only primary cause of poor fat utilization in young birds (Gomez and Polin, 1976). Lipase activity is low in the digestive tract of young birds (Noy and Sklan, 1995) but little research has been completed to examine the role of this enzyme in low fat utilization. In a series of *in vitro* studies, mammalian, fungal and bacterial lipase sources were tested under conditions which approximate the proventriculus and small intestine of young birds (Kermanshahi *et al*., 1998a, b). From these studies it was concluded that *Pseudomonas sp*. lipase has a potential to improve fat utilization in

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young birds. Therefore, the objective of this study was to investigate the role of dietary microbial *Pseudomonas sp*. lipase under in vivo conditions of young broiler chicks.

Materials and Methods:

300 day-old broiler chicks were housed in battery cages with 10 chicks/replicate and 6 cages/trearment. A liquid form of *Pseudomonas sp*. lipase (PL) was added to a corn-soybean based diet containing 8% tallow at 5 levels of 0, 50, 100, 200 and 400 units/g of diet. Feed and water provided *ad. libitum* for 2 periods of 1-21 and 21-35 days of age. Chromic oxide was used as marker. At the end of each week, 2 birds from each replicate were killed and duodenal + jejunal contents (D + J) were collected, freeze dried and 0.5 g of dried sample was mixed with 5 ml of ice cold distilled water and vortexed for 5 seconds. The vials were then centrifuged for 10 min. at 2000 rpm and 4 C and the supernatant collected in 1 ml vials and then lipase activity was measured at pH7 using tributyrin as substrate (Borgstrum and Erlanson-Albertson, 1973). Feed and feces were collected at each week, twice/d at 5-7, 12-14, 19-21 and 33-35 d of age. Fat extracted by Goldfish apparatus. Fatty acid profile of individual samples were measured by GC after esterification.

Results and Discussion:

A negative effect of dietary lipase on body weight gain, feed intake and feed to gain ratio was seen in all ages (Fig. 1).

A similar trend for fat utilization was also seen in all ages (Tab. 1).

Supplemental PL addition caused a quadratic increase in lipase activity of D + J contents at 14 d but at 21 and 35 d, a linear decrease was observed (Tab. 2). The apparent digestibility of (C18:0) was improved by the addition of PL while that of C18:1 was decreased (Tab. 3). Analysis of feeds containing 0 and 400 units/g of diet revealed that triglycerides (TG) had been nearly totally hydrolyzed to free fatty acids (FFAs) during feed mixing and/or storage in the PL supplement treatment explaining the poor performance of birds receiving FFAs (Vila and Garcia, 1996). The effect of dietary enzyme supplementation on lipase activity in the small intestine demonstrates that the dietary source in fact contributing to the supply of enzyme class in the small intestine of young broilers. However, at older ages, dietary lipase caused a decrease in digestive tract lipase activity. In theory, feeding FFAs decreases the pH of intestinal contents which could have an important impact on digestion and utilization of fat. Reducing the pH in the small intestine could decrease the activity of lipase and/or result in bile salt precipitation (DiMagno, 1982). Therefore, it can be speculated that even a minimal reduction in the secretion of bile acids, if coupled with intraluminal acidic precipitation of bile salts, could lead to fat malabsorption. Better use of C18:0 may indicate insufficient endigenous lipase in young birds and the C18:1 that the non-specific nature of PL (Jaerger *et al.*, 1994) hydrolyzes the fatty acids in 2 positions of the TG thereby reducing its utilization.

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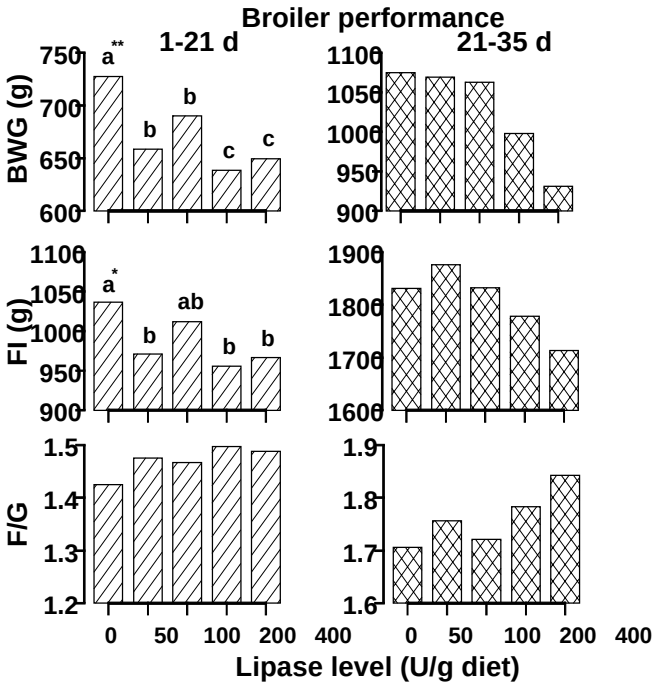

Figure 1. The effect of dietary Pseudomonas sp. lipase on body weight gain (BWG), feed intake (FI) and feed to gain ratio (F/G) from 1-21 and 21-35 days of age. Values for each parameter with different letters are significantly different (, P<0.05; **, P<0.01).*

Table 1. *The effect of dietary* Pseudomonas sp. *lipase on the apparent fat digestibility in broiler chickens*

	Fat digestibility (%)[2]		
Treatment[1]	5-7 d	19-21 d	33-35 d
0	42.7	83.5[a]	89.4[a]
50	34.5	72.0[b]	86.1[bc]
100	31.3	71.8[b]	87.8[ab]
200	35.7	72.8[b]	86.2[bc]
400	36.8	65.6[b]	84.8[c]
SEM	4.00	2.88	0.92
Probability	0.375	0.004	0.032
Contrast	----------	(P value)	----------
Regression			
Linear	0.7086	0.0014	0.0057
Quadratic	0.1275	0.2758	0.4981

[1] Supplemental lipase, units/g of diet.. [a-c] Means in columns with different superscripts are significantly different(P<0.05).. [2]Apparent fat digestibility calculated from the formula (AD=100 - (% marker in feed / % marker in feces) x (% nutrient in feces / %nutrient in feed) x 100)) as described by Saha and Gilbreath (1993).

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Table 2. *The effect of dietary* Pseudomonas sp.*on the lipase activity of pooled duodenal and jejunal contents in broiler chickens*

	Lipase activity[2]			
Treatment[1]	7 d	14 d	21 d	35 d
0	364.4	580.5	563.5[a]	667.1
50	383.8	662.5	551.9[a]	561.7
100	393.3	741.0	418.8[b]	638.5
200	407.6	700.2	513.4[ab]	486.7
400	353.8	647.0	405.4[b]	435.2
SEM	47.55	37.79	38.84	67.22
Probability	0.932	0.066	0.022	0.114
Contrast	----------		(P value)	----------
Regression				
Linear	0.7988	0.5972	0.0131	0.0175
Quadratic	0.3917	0.0124	0.6807	0.6656

[1] Supplemental lipase, units/g of diet.. [2] Micro mol free fatty acids released/min/g dry matter of digesta.. [a,b] Means in columns with different superscripts are significantly different(P<0.05)..

Table 3*. The effect of dietary* Pseudomonas sp. *lipase on the digestibility of individual fatty acids in broilers (%)*[1]

	5-7 d		19-21 d		33-35 d	
Fatty acids	Control	400 U/g diet	Control	400 U/g diet	Control	400 U/g diet
C14:0	94.79	56.94	93.01	64.80	91.15	73.60
C16:0	82.73	62.42	71.16	76.04	68.53	76.22
C16:1	39.98	41.85	59.29	38.63	70.57	60.68
C18:0	60.86	69.62	58.13	74.59	57.83	72.14
C18:1	59.85	63.71	68.55	56.36	76.72	66.61
C18:2	57.97	68.12	43.39	72.77	67.42	71.74
C18:3	71.41	63.92	72.89	68.72	87.86	50.57

[1]Apparent digestibility of individual fatty acids calculated from the formula (AD=100 - (% marker in feed / % marker in feces) x (% nutrient in feces / % nutrient in feed) x 100)) as described by Saha and Gilbreath, 1993).

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Conclusions:

Dietary supplementation with liquid form of PL caused a decrease in the performance of broilers. It is hypothesized that the negative effect of this lipase is due to the hydrolysis of TG to FFAs (even in the feed) which in turn reduces fat utilization.. The results suggest that for a lipase to be suitable for use as a dietary supplement, it shouldn't be active in feed and shouldn't hydrolyze TG completely to FFAs. Questions are raised to whether the form, dosage or specificity of lipase application has an impact on lipid hydrolysis and its utilization? Further research is needed to answer to these questions.

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