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Effect of Oral Supplementation of Putrescine and L-glutamine on the Histomorphology of Small Intestine and Growth Performance in Broiler Chickens

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ABSTRACT

A work was conducted to study the effect of putrescine and L-glutamine supplementation on histomorphology of small intestine in broiler chickens. Five groups of ten chicks each in three replicates reared up to 42 days were used for this study. Putrescine and L-glutamine were orally supplemented at 0% level as control (T1), putrescine 0.05% (T2), putrescine 0.1% (T3), L-glutamine 0.5% (T4) and L-glutamine 1% (T5) from 0 day to 7 days after hatch. At 2nd and 3rd week of age, 3 birds from each groupwere slaughtered and tissue samples from small intestine (duodenum, jejunum and ileum) were collected for histomorphological studies. Statistically significant ($p<0.01$) increase in duodenal and jejunal villi length, width,crypt length and ileal villi length and width were observed in L-glutamine fed groups than putrescine and control groups. There was no significant difference in duodenal crypt length/width, jejunal crypt width and ileal crypt length/width. It can be inferred that L-glutamine promoted the intestinal villi development during early posthatch life, thus increasing the nutrients absorption and growth performance as body weight gain in broiler chickens.

Keywords: L-Glutamine, Putrescine, chicken, Histomorphology, Growth performance.

Functional development of the intestines is essential for successful poultry production. During the early days post hatch, the rate of development of the small intestine exceeds the rate of body weight gain (Sklan, 2001). In poultry production practices, long delays in access to feed and water until placement of chicks in the farm are common for broiler chickens and turkeys because newly hatched birds are removed from the hatchery only after almost all eggs have hatched (Moran, 2007). The egg supplies nutrients during embryonic development, and after hatching, the birds start to have exogenous diets. This change necessitates an adaptation period for the gastrointestinal tract of the birds, because the GIT undergoes a post hatch maturation process that can significantly affect performance, mainly in the first 2 weeks post hatch, which represents approximately 30 per cent of the useful life of the broiler bird. The development of gastrointestinal tract during the period posthatch has a major role in inducing growth (Nir *et al*., 1993). The small intestine mucosal growth is stimulated when putrescine or ethylamine was infused in to the small intestine, or when dimethylamine was fed to rats Dembinski *et al.*, 1984; Seidel *et al.*, 1985).

The present work was planned to study the effect of putrescine or glutamine on the histomorphology of post hatch broiler chicken.

MATERIALS AND METHODS

One hundred and fifty straight run day old broiler chicks (Vencobb 400) were wing banded and randomly allotted to five groups with three replicates of ten chicks based on their body weight and cage reared under standard management practices up to 42 days of age. Putrescine and L-Glutamine were orally supplemented along with standard diet at 0% level as Control (T1), putrescine 0.05% (T2), putrescine 0.1% (T3), L-glutamine 0.5%

(T4) and L-glutamine 1% (T5) from 0 day to 7 days after hatch. Broiler prestarter, starter and finisher diets were formulated according to standards of BIS (1992) and fed *ad libitum* to the birds from 1 to 12, 12 to 22 and 22 to 42 days of age, respectively. The broiler birds were vaccinated against Ranikhet disease on day 7 and 21, and Infectious Bursal Disease at the age of day 14. At the end of every week, body weight of individual bird was recorded and body weight gain was calculated.

At the end of the second and third week, three birds were slaughtered and samples from the duodenum, jejunum and ileum (each ~ 2.5 cm in length) were collected, fixed, processed and embedded in paraffin and sectioned to 3-4 µm thickness and stained with haematoxylin and eosin for histomorphological studies (Bancroft and Stevens, 1996). The micrometrical measurements (villi length,width and crypt length, width were taken from all the three parts of the small intestine by using Leica histological image analyser software.

Statistical analyses were done using completely randomized design (CRD) as prescribed by Snedecor and Cochran (1994).

RESULTS AND DISCUSSION

The mean body weight at 6th week in 0.5% glutamine (T4) group was significantly (P<0.05) higher than T3, T5 and control. (Table1). At first week of age a significant (P<0.01) reduction in body weight was observed in 0.1% putrescine (T3) group compared to other groups.

The findings of the present study agrees with Smith (1990a), Sousadias and Smith (1995) and Yi *et al*. (2005) who reported better body weight in broilers fed putrescine and L-glutamine. The enhanced weight gain in T4 group might be due to the effect of glutamine on small intestinal villi length and width (Table 1) as reported by Girdhar *et al*. (2006) and Murakami *et al*. (2007) who reported that supplementing dietary putrescine and L-glutamine increased small intestine villus height, villus width, crypt depth and thickness of the sub mucosa in turkey poult and broiler chickens.

The lowest body weight recorded in T3 at first week of age might be due to toxicity of putrescine. Smith (1990b) observed that chicks fed 0.2 per cent putrescine had improved growth. But in the present study 0.1 per cent putrescine itself depressed the body weight of broiler chickens.

A significant (P< 0.01) increase in duodenal villi length was observed in 0.5 and 1 per cent glutamine fed chicks compared to putrescine groups and control during second week of age (Table 2). Similarly, T3, T4 and T5 groups had higher villi length compared to T2 and control groups during 3rd week of age. However, duodenal villi width, crypt length and crypt width did not differ between the groups during second and third weeks of age

Jejunal villi length and width were significantly improved in all treatment groups during second (P< 0.05) and third (P< 0.01) week of age compared to control. Similarly, crypt length was higher in all the treatment groups compared to control at second week of age. No significant difference was observed in crypt width between groups at second and third weeks of age.

Mean ileal villi length was significantly (P< 0.01) higher in glutamine fed groups compared to putrescine and control groups at 2nd weeks of age. Putrescine did not improve ileal villi length as that of glutamine at second week of age. At third week of age, villi length was significantly (P<0.01) higher in all the treatment groups compared to control. Similarly, villi width was greater in T3, T4 and T5 compared to T2 and control groups at second week of age and by third week of age, all the treatment groups had significantly (P<0.01) higher width than control. The crypt length did not differ between groups during second and third week of age.Whereas crypt width was significantly (P<0.01) higher in T4 group compared to control.

Observations of the present study agreed with Murakami *et al*. (2007) who observed that 1 per cent glutamine in the starter diet for the first 7 or 14 day of life with 1 mg of Vitamin E/kg caused greater development in the duodenum followed by the jejunum and ileum at 41 day of life. Fischer da Silva *et al*. (2007) also observed that birds supplemented with 1 per cent glutamine and fed *ad libitum* showed larger surface area in the enterocytes of the jejunum and ileum at 21 day old birds.Additionally,Jazideh *et al*., (2014) reported that 0.5 % glutamime in the diet improved the intestinal morphology and consequently increased the body weight gain in broiler chickens.

In conclusion, results of this study suggested that the L-glutamine increased the length and width of the micro

villi of the small intestine, suggesting more surface area for absorption at the tip of the enterocytes in the small intestine of broilers, which in turn increases the weight gain in broilers chickens.

Table 1. Mean (±SE) body weight (g) of broiler chickens fed different levels of putrescine and L-glutamine from 1 to 7 days of age

Treatment	Age in weeks					
	I	II	III	IV	V	VI
T_1- Control	174.72^{c}±3.12	380.61±14.07	715.00±16.71	1201.11^{b}±38.28	1769.33^{a}±31.67	2097.22^{b}±41.75
T_2- 0.05% put	146.67^{ab}±4.47	364.72±9.79	682.22±23.39	1121.61^{a}±25.09	1714.39^{a}±30.15	2204.45^{ab}±38.91
T_3- 0.1% put	136.61^{a}±4.80	342.50±9.46	676.45±10.61	1077.22^{a}±28.29	1589.10^{b}±46.08	2204.89^{b}±49.98
T_4- 0.5% L-glu	176.39^{c}±3.26	356.06±19.74	737.22±19.75	1198.89^{b}±21.85	1769.72^{a}±30.44	2325.00^{a}±31.34
T_5- 1 % L-glu	162.11^{bc}±5.41	366.83±5.49	713.89±11.84	1191.11^{b}±16.64	1739.88^{a}±50.48	2190.00 ^{b}±42.80

Mean of 6 observations. Means bearing lower case letters in a column differ significantly (P< 0.05).
Put = Putrescine
L-glu = L-glutamine

Table 2. Mean (±SE) villi length (µm) and width (µm) of duodenum, jejunum and ileum at 2nd and 3rd week of age in broiler chickens fed different levels of putrescine and L-glutamine from 1 to 7 days of age

Treat Ment	Villi length						Villi width					
	Second week			Third week			Second week			Third week		
	Duodenum	Jejunum	Ileum	Duodenum	Jejunum	ileum	Duodenum	Jejunum	ileum	Duodenum	Jejunum	ileum
T_1-Control	963.23^{c}	762.12^{b}	598.77^{b}	1115.38^{b}	1027.12^{b}	768.81^{b}	103.22	73.03^{b}	43.72^{b}	114.41	78.87^{a}	62.15^{a}
	±26.99	±20.66	±26.96	±26.65	±27.50	±32.17	±5.66	± 4.12	± 2.33	±2.30	± 2.12	±2.50
T_2 - 0.05% put	1142.73^{b}	875.82^{a}	676.08 ^{b}	1154.26^{b}	1173.04^{a}	911.23^{a}	114.78	94.256^{a}	39.695^{c}	111.56	85.80^{a}	79.15^{b}
	±28.92	±42.39	±21.14	±6.7	±21.97	±34.26	±6.7	±4.23	±3.17	±2.32	±1.51	±2.50
T_3 - 0.1% put	998.43^{c}	849.14^{a}	684.62^{b}	1306.32^{a}	1189.32^{a}	894.90^{a}	116.37	99.59^{a}	67.50^{a}	114.81	85.16^{a}	79.49^{b}
	±9.09	± 2.39	±30.23.	±5.65	±41.58	± 9.46	±5.65	± 5.22	± .31	±1.01	±1.24	± 1.84
T_4 - 0.5%L glu	1346.03^{a}	902.40^{a}	802.91^{a}	1268.49^{a}	1237.65^{a}	926.70^{a}	121.01	89.49^{a}	69.56^{a}	112.64	89.14^{b}	84.99^{b}
	±50.16	± 23.02	±17.15	±2.85	±38.97	±30.54	±2.85	±4.80	±5.17	±2.52	± 2.53	± 3.79
T5 - 1% L glu	1322.72^{a}	876.82^{a}	831.69^{a}	1407.93^{a}	1198.29^{a}	948.99^{a}	113.56	99.54^{a}	75.32^{a}	113.72	90.30^{b}	88.49^{b}
	±55.10	±26.89	± 40.30	±4.66	± 7.86	± 30.23	±4.66	± 3.82	± 3.04	±2.78	± 1.95	±3.04

Mean of 6 observations. Means bearing different superscript letters within a column differ significantly (P< 0.05)
Put = Putrescine L-glu = L-glutamine

Table 3. Mean (± SE) crypt length (µm) and width (µm) of duodenum, jejunum and ileum at 2nd and 3rd week of age in broiler chickens fed different levels of putrescine and L- glutamine from 1 to 7 days of age

Treatment	crypt length						crypt width					
	Second week			Third week			Second week			Third week		
	Duodenum	Jejunum	ileum	Duodenum	Jejunum	ileum	Duodenum	Jejunum	ileum	Duodenum	Jejunum	ileum
T1-Control	963.23^{c}	762.12^{b}	598.77^{b}	1115.38^{b}	1027.12^{b}	768.81^{b}	103.22	73.03^{b}	43.72^{b}	114.41	78.87^{a}	62.15^{a}
	±26.99	±20.66	±26.96	±26.65	±27.50	±32.17	±5.66	± 4.12	± 2.33	±2.30	± 2.12	±2.50
T2 - 0.05% put	1142.73^{b}	875.82^{a}	676.08 ^{b} ±21.14	1154.26^{b}	1173.04^{a}	911.23^{a}	114.78	94.256^{a}	39.695^{c}	111.56	85.80^{a}	79.15^{b}
	±28.92	±42.39		±6.7	±21.97	±34.26	±6.7	±4.23	±3.17	±2.32	±1.51	±2.50
T3 - 0.1% put	998.43^{c}	849.14^{a}	684.62^{b}	1306.32^{a}	1189.32^{a}	894.90^{a}	116.37	99.59^{a}	67.50^{a}	114.81	85.16^{a}	79.49^{b}
	±9.09	± 2.39	±30.23.	±5.65	±41.58	± 9.46	±5.65	± 5.22	± .31	±1.01	±1.24	± 1.84
T4 - 0.5%L glu	74.68	44.20^{a}	91.86	88.13	53.96	114.70	30.61	23.34	28.37	36.52	34.09	46.32^{a}
	±1.97	± 3.72	± 3.88	±1.85	± 3.87	± 5.12	±2.56	± 2.87	± 1.66	±2.62	± 2.73	± 4.44
T5 - 1% L glu	78.72	58.47^{b}	80.78	89.30	57.37	112.19	29.53	33.63	35.82	37.14	37.94	39.11^{ab}
	±3.23	± 6.41	± 5.90	±2.08	± 5.78	± 4.86	±1.77	± 2.62	± 4.71	±2.05	± 1.43	±1.88

Mean of 6 observations. Means bearing different superscript letters within a column differ significantly (P< 0.05)

Put = Putrescine L-glu = L-glutamine

SUMMARY

There was a positive influence of both putrescine and L-glutamine supplementation on intestinal morphology was observed. The villi length, increased significantly ($p<0.05$) in a dose related manner among the treatment groups than control group at both second and third week of age. There was no significant ($p<0.01$) difference in villi width and crypt width at both second and third week of age studied. This increased villi morphology accelerate the surface area of small intestine for absorption of nutrient and inturn increased the final body weight.

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