

Effect of Protein Synthesis Modulator and Acute Heat Stress on Jejunal Morphology in Broiler Chicken

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Abstract

An experiment was conducted to assess the effect of protein synthesis modulator and heat stress on jejunal morphology in broiler chicken. A total one hundred ninety two of CARI-BRO Vishal broiler chicken were reared up to five weeks of age under standard managemental conditions and 36th day of age divided into three treatments including control, enhancer and inhibitor, receiving intra peritoneal normal saline, (0.5 ml) glutamine (0.75 mg kg⁻¹ of BW) and quercetin (5 mg kg⁻¹ of BW) respectively. After, 24 hours later each they were exposed to different duration of acute heat stress for 0, 2, 5 and 10 hours under 40±1 °C; 55% RH in psychometric chamber, after exposure of heat stress birds are immediately sacrificed (n=4 for each exposure duration) and jejunum tissue sample were collected in 10% formal saline and further processed for morphological study. A significant ($p < 0.05$) effect of protein synthesis modulator on jejunal morphological parameters was observed in broilers exposed to varied periods of heat stress. Protein synthesis modulator revealed a significant effect ($p < 0.05$) on villi length of jejunum however heat stress exposure and duration of heat stress did not show any significant effect ($p > 0.05$) on jejunal morphological observations as compared to normal villi length, crypt depth and villi length to crypt depth ratio. It was concluded that protein synthesis modulator glutamine protect intestinal morphology under heat stress condition.

1. Introduction

The present scenario of changing climate with increasing environmental temperature, heat stress is considered to be the major cause of loss of production and reduced profit in the poultry production worldwide (Sejian et al., 2012). Commercial broiler chickens more susceptible to exposure of heat stress because of higher, growth rate and metabolic heat production, while heat dissipation does not. Heat stress exerted negative effect on feed intake, body weight gain (Geraert et al., 1996) and increase mortality rates (Smith, 1993). During thermal stress intestinal tissue are most affected because of its major role in body barrier, digestion and absorption of nutrient and immunity. Heat stress induces increase in intestinal permeability that associated with massive generalized sloughing of small intestinal epithelial layer from the villus tips and lysis of intestinal epithelial cells, indicating that the increase in intestinal permeability due to the extensive damage of the epithelial surface (Lambert, 2009).

Protein synthesis modulator Glutamine is the non-essential nutrient but under heat stress condition glutamine act as conditioned essential nutrient (Murakami et al., 2007) that protect intestinal epithelium by enhancing heat shock protein (Wischmeyer et al., 1997). Supplementation of glutamine in diet, that preserved gut integrity in rodents (Larson et al., 2007) and pigs (Wu et al., 1996) and in broiler chicken it improved gastrointestinal tract development (Soltan, 2009). Another Protein synthesis modulator quercetin that is bioflavonoid (Hosokawa et al., 1992) used as a negative control in our experiment. Hence, the current study was designed to investigate the effect of protein synthesis modulator, glutamine and acute heat stress on jejunal morphology in broiler chicken.

2. Materials and Methods

A total of one hundred ninety two CARI-BRO Vishal broiler chickens with similar body weight were housed in cages and reared up to five weeks from March to April 2015 under



standard management conditions. On 36th day divided into 6 groups with three treatments (32 birds in each group's) including control, enhancer and inhibitor, receiving intra peritoneal normal saline, glutamine @ 0.75 mg kg⁻¹ body weight and Quercetin @ 5 mg kg⁻¹ body weight respectively. After, 24 hours of administration each of the six groups were again divided into two with three groups in each, one being exposed to acute heat stress (40±1 °C; 55% RH) for different duration of 0, 2, 5 and 10 hours in psychometric chamber and another one were kept as unexposed to heat stress. After exposure of different duration of heat stress (n=4 for each exposure duration) killed by cervical dislocation and longitudinal sections of jejunum tissue samples were collected and fixed in 10% formal saline. The fixed samples were cut into pieces of 2–3 mm thickness and washed thoroughly in tap water overnight before dehydrating the tissues in ascending grades of alcohol (50%, 60%, 70%, 80%, 90% absolute alcohol

I and II). The dehydrated tissues were cleared in benzene and embedded in paraffin blocks. Serial section of 5 µ thickness were cut and stained with hematoxylin and eosin (Culling, 1968). Villus height and crypt depth were measured under a light microscope. A total of 10 intact, well oriented, crypt-villi were selected in triplicate for jejunal cross section. Villus height was measured from its tip to villus crypt junction, and crypt depth was measured as the depth of invagination between adjacent villi. The data obtained from experiment were analysed by 2×3×4 factorial design using SPSS V.20 for both interaction and main effect. The means were compared using Tukey test.

3. Results and Discussion

In the present study interaction and main effect of protein synthesis modulator on jejunal morphological parameters (Mean values±SE) are represented in Table 1 and 2. Results

Table 1: Effect of hatching temperature and protein synthesis modulator at different periods of heat stress exposure on jejunal morphological parameters

Heat stress	Protein modulator	Hours	Villi length (μm)	Crypt depth (μm)	Villi length:Crypt depth ratio
Unexposed	Control	0	915.03 ^a ±44.82	237.68 ^{ab} ±12.36	3.88 ^{ab} ±0.32
		2	960.10 ^{ab} ±14.81	278 ^b ±22.10	3.50 ^a ±0.30
		5	998.87 ^{ab} ±54.14	237 ^{ab} ±38.13	4.53 ^{ab} ±1.02
		10	1011.07 ^{ab} ±40.20	181.37 ^{ab} ±17.04	5.64 ^{ab} ±0.36
	Enhancer	0	1012.77 ^{ab} ±15.75	247.20 ^{ab} ±27.04	4.19 ^{ab} ±0.41
		2	1000.37 ^{ab} ±33.99	158.57 ^a ±17.10	6.45 ^b ±0.72
		5	1017.70 ^{ab} ±46.76	178.83 ^{ab} ±4.19	5.69 ^{ab} ±0.24
		10	1096.83 ^{ab} ±41.24	215.37 ^{ab} ±3.81	4.99 ^{ab} ±0.28
	Inhibitor	0	1023.33 ^{ab} ±34.99	194.10 ^{ab} ±12.19	5.29 ^{ab} ±0.14
		2	1073.75 ^{ab} ±22.41	200.14 ^{ab} ±16.86	5.42 ^{ab} ±0.35
		5	1089.87 ^{ab} ±52.20	239.47 ^{ab} ±22.59	4.67 ^{ab} ±0.65
		10	1110.33 ^{ab} ±42.09	226.40 ^{ab} ±13.42	4.96 ^{ab} ±0.49
Exposed	Control	0	982.53 ^{ab} ±4.68	240.07 ^{ab} ±15.69	4.13 ^{ab} ±0.28
		2	1055.93 ^{ab} ±57.86	207.70 ^{ab} ±5.54	5.09 ^{ab} ±0.32
		5	1082.93 ^{ab} ±60.79	221.77 ^{ab} ±37.51	5.19 ^{ab} ±0.91
		10	934.03 ^{ab} ±29.80	222.83 ^{ab} ±21.18	4.24 ^{ab} ±0.29
	Enhancer	0	1051.37 ^{ab} ±28.98	226.20 ^{ab} ±20.91	4.74 ^{ab} ±0.50
		2	1052.27 ^{ab} ±28.57	246.90 ^{ab} ±19.64	4.31 ^{ab} ±0.29
		5	980.00 ^{ab} ±24.15	189.27 ^{ab} ±3.80	5.19 ^{ab} ±0.22
		10	1050.20 ^{ab} ±19.27	222.70 ^{ab} ±27.02	4.86 ^{ab} ±0.61
	Inhibitor	0	1061.23 ^{ab} ±60.36	217.47 ^{ab} ±24.38	5.05 ^{ab} ±0.77
		2	1006.93 ^{ab} ±31.75	242.23 ^{ab} ±19.63	4.24 ^{ab} ±0.50
		5	995.63 ^{ab} ±20.52	197.32 ^{ab} ±16.57	5.10 ^{ab} ±0.32
		10	1032.77 ^{ab} ±26.79	212.87 ^{ab} ±21.73	4.97 ^{ab} ±0.57
<i>p</i> value			0.000	0.000	0.000

^{ab}Mean values bearing different superscripts within columns differ significantly (**p*<0.05)



Table 2: Effect of protein synthesis modulator on jejunal morphological parameters

Protein synthesis modulator	Villi length	Crypt depth	Villi length: crypt depth Ratio
Control	992.56 ^a ±10.27	228.30±8.74	4.52±0.14
Enhancer	1032.68 ^{ab} ±9.45	210.62±6.54	4.96±0.14
Inhibitor	1049.23 ^b ±10.53	216.24±5.96	5.05 ±0.13
<i>p</i> -value	0.010	0.219	0.094

^{ab}Mean values bearing different superscripts within columns differ significantly ($*p<0.05$)

indicated significant ($*p<0.05$) effect of protein synthesis modulator on jejunal morphological parameters was observed in broilers exposed to varied periods of heat stress and Protein synthesis modulator revealed a significant effect ($*p<0.05$) on villi length in jejunum. Inhibitor group showed an increased villi length as compared to control and enhancer groups. Enhancer significantly improves the jejunal morphology; increase in villus height was observed as compared with control. Result of present study in agreement with the finding of Olubodun et al. (2015) who indicated that 1% glutamine supplementation to broiler chicken under hot and humid conditions improves intestinal morphology. Hence, there is a possibility that the protein modulator, increase in villus height it may enhance nutrient absorption by increasing the intestinal surface area (Bartell and Batal, 2007; Soltan, 2009; Moghaddam et al., 2013). Another study in rats showed that oral glutamine decrease intestinal permeability and improves survival from hyperthermia injury by enhancing heat shock protein expression, (Singleton and Wischmeyer, 2006). Wischmeyer (2002); Zulkifli et al. (2003) also noticed similar findings. The increased villi height in the inhibitor group obtained in the present experiment it might be due to the compensatory mechanisms of the bird to adjust its digestive absorption process under heat stress condition. Negative effect of heat stress on feed intake and feed conversion ratio is due to the damage to the intestinal mucosal structure and digestion absorption function (Ryder et al., 2004). Studies of Liu et al. (2009); Yu et al. (2010) found that heat stress caused noticeable damage to porcine intestinal epithelia, which included injury to the tips of the intestinal villi, inducing epithelial cell shedding, exposing the intestinal mucosa lamina propria, as well as reduction in the size of villus height and crypt depth. Ning et al. (2003) reported that there were harsh effects of heat stress on pathological damage of the duodenum, jejunum, and ileum, which mainly involved mucosal epithelial cell exfoliation and villi fracture. In present experiment Mean±SE values relating to effect of heat stress exposure and duration on jejunal morphological observations is presented in Table 3 and 4 respectively. Heat stress exposure did not show any significant effect ($*p<0.05$) on villi length, crypt depth and villi

Table 3: Effect of heat stress exposure on jejunal morphological parameters

Heat stress	Villi length	Crypt depth	Villi length :Crypt depth ratio
Unexposed	1037.33±9.27	217.65±7.25	4.93±0.12
Exposed	1031.72±7.61	192.52±5.81	4.94±0.11
<i>p</i> -value	0.893	0.597	0.394

Table 4: Effect of duration of heat stress exposure on jejunal morphological parameters

Duration of heat stress	Villi length	Crypt depth	Villi length : Crypt depth ratio
0 hour	1017.71±13.44	227.11±6.18	4.54±0.14
2 hours	1024.89±11.43	222.25±7.58	4.83±0.18
5 hours	1027.50±10.16	210.60±7.01	4.94±0.16
10 hours	1039.20±12.22	213.58±5.74	5.06±0.17
<i>p</i> -value	0.524	0.479	0.337

length to crypt depth ratio. Similarly duration of heat stress also did not pose any significant effect ($p>0.05$) on jejunal morphological observations as compared to normal. We did not find significant changes in the jejunal morphology among the birds under heat stress in our study. Results of our study are in agreement with the finding of Hao et al. (2012) who reported that no significant changes on intestinal morphology in response to heat stress. These discrepancies might be due to different temperatures and different duration heat stress. However, further more studies are required for establishing the relationship the jejunal morphology with respect to acute heat stress.

4. Conclusion

Protein synthesis modulator glutamine protect intestinal morphology under heat stress condition.

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