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Comparison of Intestinal Response of Chicks from Light and Heavy Eggs to Posthatch Fasting

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Abstract: The effects of posthatch fasting on villi height and number, crypt depth and number of globet cells in duodenum, jejunum and ileum of broiler chicks from heavy and light eggs were compared. The 2 x 3 x 3 factorial design (egg weight: light and heavy eggs; treatments: with water and feed, with water, without water and feed; treatment duration: 24, 48 and 72 h) was used. The villi presented higher size in chicks from heavy than from light eggs. The fasting resulted in lower villi in duodenum (at 48 h), jejunum and ileum (at 72 h). The villi number increased in duodenum and jejunum of chicks from light eggs and only in jejunum of chicks from heavy eggs, but the increase was more accented in chicks from light ones. The fasting reduced the globet cells number in jejunum. Water intake avoided the fasting effects on villi height but had no effect on villi number. Chicks from heavy eggs fed with water and ration presented deeper crypts in all regions of the small intestine. The duodenum and ileum crypt depth of the chicks from heavy eggs reduced when they were submitted to fasting and when they were fed only with water. The results showed that chicks from light eggs were more affected than chicks from heavy eggs. The water intake partially avoided the fasting effects.

Key words: Globet cells, crypt depth, egg weight, posthatch fasting, villi height, villi number

INTRODUCTION

The delayed access of the chicks to feed after hatching results in loss from 5-10% of body weight (Baião and Cançado, 1998), in slow development of intestinal villi (Shamoto and Yamauchi, 2000), high villi density (Maiorka *et al*., 2002, 2003), epithelial cells death (Yamauchi *et al*., 1996), cell extrusion (Gomide *et al*., 2004) and hematological alterations (Pires *et al*., 2007). This posthatch fasting delays the broiler growth (Nir and Levenon, 1993; Almeida, 2002), hindering the weight gain and decreasing the broiler uniformity at slaughter age.

It is known that chicks from young breeders present lower body weight at hatch and posthatch period than those from old breeders (Noy and Pinchasov, 1993). These differences have been attributed to the lower quantity of albumen and yolk in eggs produced by young breeders, as well as to the low proportion of water and lipid (May and Stadelman, 1960; McNaughton *et al*., 1978) and low protein concentration (Cardozo *et al*., 2002).

The results cited above arise the question if the effects of posthatch fasting on chicks from light and heavy eggs are similar. According to Riccardi *et al*. (2009), the fasting of feed and water or only feed induces more accented reduction in liver weight of chicks from light eggs than those from heavy ones, even promoting faster absorption of yolk sac in chicks from light eggs.

Considering the importance of the intestinal development to broiler posthatch growth and that the adequate handling of the chicks can diminish economic losses in domestic avian production, the objective of the present experiment was to evaluate the effect of the posthatch fasting of feed and water or only feed at 24, 48 and 72 h on intestinal mucosa of chicks from light and heavy eggs.

MATERIALS AND METHODS

The fertile eggs were obtained from a commercial hatchery when broiler breeders (Cobb®) were 29-week-old (62±1.8 g: light eggs) and 45-week-old (74±2.6 g: heavy eggs). The eggs were incubated at 37.5°C and 60% of humidity in forced-air rotating incubators (IP120, Premium Ecológica) with automatic control of turnings (1 turn/2 h) and temperature.

After hatching, the male chicks were homogeneously separated in six groups (two groups of 45 chicks for each treatment), according to the body weight and were submitted to the following treatments: free access to water and ration (2,800 kcal/Kg and 22% of protein), free access to water and fasting of feed and water. A total of 5 chicks per treatment at 24, 48 and 72 h after the

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beginning of the treatments were randomly selected and used for small intestine analyses. All groups were bred in commercial cages (Premium Ecológica, 50 cm x 90 cm), containing two 40W light bulbs (one green and another red). The chicks used in this experiment were hatched within a period of 3 h.

Samples (from 1 to 2 cm) were taken from the proximal half of the duodenum, from the region immediately before Meckel`s diverticulum (jejunum) and from the region immediately before ileocecal junction (ileum). All tissue samples were longitudinally opened on a piece of paper, rapidly and gently washed with water and immediately fixed in Bouin solution for 24 h at quarter of temperature. After washing with 50% alcohol, the samples were dehydrated in graded ethanol series (70, 80, 90 and 100%), cleared in xilol (100%, 2 times) and embebbed in paraffin. Ten sections per intestinal segment per bird were cut (6 µm), placed on glass slides, deparaffinized and stained with PAS. The analyzed parameters were the villi height and crypt depth (mm, 30 measurements per intestinal segment per bird) and the number of villi and globet cells. The villi number was counted along 205 µm of intestinal mucosa sections (10 counts per intestinal segment per bird). The number of globet cells PAS^+ was counted along 410 µm of villus epithelium (10 counts per segment per bird). The data were obtained using image analyzed systems linked to a microscopy or stereomicroscopy (Leica).

The completely randomized design was used with the following variables: 2 egg weights (light and heavy eggs), 3 treatment types (water and ration supply, water supply and fasting of water and feed) and 3 treatment durations (24, 48 and 72 h). Data were submitted to analysis of variance and the pair-wise comparisons of the means were made using Tukey's test procedure. A P-value $\leq$0.05 was considered significant. All of the statistical results were obtained from SAS software package (2004).

The protocol of the experiment was approved by the Committee of Ethics in Animal Use (CEUA) of the Faculty of Agricultural and Veterinary Sciences (protocol number 025055/10).

RESULTS

Duodenum: The Villi Height (VH) in this intestinal segment was significantly ($p\leq0.05$) influenced by egg weight, type and duration of the treatments and there was significant interaction ($p\leq0.05$) between treatment type and duration (Table 1). The VH was higher for chicks from heavy than for light eggs and for chicks fed than for chicks submitted to fasting of water and feed, increasing with the age (Table 1). According to the mentioned interaction (Table 4), differences in VH between fed and fasted chicks occurred from 48 h. Besides, temporal comparisons show that the duodenum VH increased from 24-72 h for chicks with access to water and ration and from 24-48 h for chicks with access only to water, but did not increase for chicks submitted to fasting.

The Duodenal Villi Number (DVN) was significantly ($p\leq0.05$) influenced by type and duration of the treatments and there was significant interaction ($p\leq0.005$) between egg weight and treatment type. The DVN was higher in chicks with access to water than in fed chicks and fasted chicks, that did not differ between themselves (Table 1). It was also observed that the DVN decreased from 24-72 h. The interaction showed at Table 5, evidenced that there were no differences in DVN for the different treatments between chicks from light and heavy eggs. Differences in the DVN among treatments were found only for chicks from light eggs, in which the smallest DVN was observed when they were fed and were not detected differences in this parameter between chicks from light eggs that were submitted to feed and water withdrawal and those that had access to water.

Table 1: Effects of egg weight and treatment type and duration on Villi Height (VH, mm) and Number (VN), Crypt Depth (CD, mm) and Globet Cells Number (GCN) in duodenum of male broiler chicks

	VH	VN	CD	GCN
Egg Weight (EW)				
Light eggs	1.19±0.39 B	7.80±0.89	0.06±0.01 B	14.39±2.73 B
Heavy eggs	1.30±0.42 A	7.90±1.05	0.07±0.01 A	18.68±3.14 A
Treatment Type (TT)				
Water + ration	1.91±0.31 A	7.02±0.83 B	0.08±0.01	16.43±3.93
Water	1.77±0.24 B	7.69±0.62 A	0.07±0.01	16.45±2.76
Fasting	1.60±0.22 C	7.23±0.66 B	0.08±0.01	15.40±3.78
Treatment Duration (TD)				
24 h	1.67±0.19 B	7.62±0.76 A	0.08±0.01 A	14.55±2.85 B
48 h	1.79±0.31 AB	7.31±0.73 AB	0.07±0.01 B	15.62±2.73 A
72 h	1.85±0.33 A	6.97±0.66 B	0.07±0.01 B	18.00±3.94 A
Probability				
EW	0.0101	0.4939	0.0001	0.0001
TT	0.0001	0.0005	0.1841	0.1151
TD	0.0073	0.0007	0.0001	0.0001
EW x TT	0.2396	0.0202	0.0001	0.0722
EW x TD	0.4320	0.7635	0.1699	0.3016
TT x TD	0.0015	0.3130	0.0010	0.0035

A-B: Means followed by different letters (columns) are significantly different ($p\leq0.05$)

Table 2: Effect of egg weight and treatment type and duration on Villi Height (VH, mm) and Number (VN), Crypt Depth (CD, mm) and Globet Cells Number (GCN) in jejunum of male broiler chicks

	VH	VN	CD	GCN
Eggs Weight (EW)				
Light eggs	0.96±0.17 B	7.99±0.99	0.07±0.01	13.74±1.69 B
Heavy eggs	1.11±0.18 A	8.00±1.02	0.07±0.01	19.35±3.41 A
Treatment Type (TT)				
Water + ration	1.07±0.21	7.23±0.82 B	0.07±0.01	17.64±4.49 A
Water	1.02±0.15	8.46±0.72 A	0.07±0.01	16.85±3.27 A
Fasting	1.02±0.19	8.32±1.02 A	0.07±0.01	15.44±3.57 B
Treatment Duration (TD)				
24 h	0.93±0.16 B	8.44±1.13 A	0.07±0.01 AB	14.87±2.64 C
48 h	1.09±0.17 A	7.76±0.84 B	0.06±0.01 B	16.84±3.40 B
72 h	1.11±0.18 A	7.68±0.87 B	0.08±0.01 A	18.62±4.60 A
Probability				
EW	0.0001	0.8588	0.1557	0.0001
TT	0.2882	0.0001	0.2180	0.0028
TD	0.0001	0.0002	0.0279	0.0001
EW x TT	0.4001	0.0109	0.0344	0.3448
EW x TD	0.1364	0.3041	0.1159	0.2376
TT x TD	0.0006	0.2306	0.2905	0.0996

A-B: Means followed by different letters (columns) are significantly different ($p \leq 0.05$)

Duodenal Crypt Depth (DCD) was significantly ($p \leq 0.05$) influenced by egg weight and treatment duration and there was significant ($p \leq 0.05$) interaction between egg weight and treatment type, as well as between type and duration of the treatments (Table 1). The duodenal crypt was deeper in chicks from heavy than from light eggs and at 24 than 48 and 72 h, that did not differ between themselves. The first interaction showed that there were no differences in the DCD among treatments along the experimental period (from 24 to 72 h) and that only chicks without access to water and ration presented decrease of DCD gradually along the period of fasting of feed and water (Table 4). The DCD of the chicks submitted to the other treatments (supply of water and ration and only water) did not present variations during the experimental period. The interaction between egg weight and treatment duration (Table 5) showed that chicks from heavy eggs presented higher DCD than chicks from light eggs when water and ration or only water were supplied, but this trend was not observed when the chicks were submitted to fasting. Besides, chicks from light eggs had deeper duodenal crypt when fasted than when fed with water and ration or only water, while chicks from heavy eggs presented higher DCD when fed than when received water or were submitted to feed and water withdrawal.

The Table 1 also shows significant ($p \leq 0.05$) effect of weight egg and treatment duration on Globet Cells Number (GCN) in duodenum. Chicks from heavy eggs presented higher GCN than those from light eggs. The GCN was greater at 72 h than at 24 and 48 h. Significant ($p < 0.05$) interaction between type and duration of the treatments (Table 1, 4) was detected, showing no differences in GCN among treatments in the 3 analyzed periods. However, fed and fasted chicks presented an increase of the GCN from 48-72 h, while this parameters remained unchanged in the chicks with supply of only water during experiment.

Jejunum: Significant effect ($p \leq 0.05$) of egg weight and treatment duration on Jejunal Villi Height (JVH) was detected, as well as significant interaction ($p \leq 0.05$) between type and duration of the treatments (Table 2). The JV was higher in chicks from heavy than those from light eggs and increased from 24-48 h. The interaction showed that fasted chicks did not present change in the JVH during the experimental period, differently from the chicks that had access to water and ration or only water, in which the values increased from 24-48 h (Table 4). The interaction also showed that there were differences among treatments only at 24 and 72 h. At 24 h, the JV was higher in fasted chicks than in fed chicks, while at 72 h the JV was higher in fed than fasted chicks (Table 4).

The Jejunal Villi Number (JVN) was influenced ($p \leq 0.05$) by type and duration of the treatments and there was significant interaction ($p < 0.05$) between egg weight and treatment duration (Table 2). The JVN was lower in fed than in fasted chicks and chicks with access to water, that did not differ between themselves. Besides, the JVN decreased from 24-48 h, maintaining the values until 72 h. The interaction showed difference between chicks from light and heavy eggs when they had access to water and ration (Table 5), being the JVN lower in chicks from light than from heavy eggs. Comparisons among the treatments demonstrated that chicks from light and heavy eggs fed with water and ration presented lower JVN when compared with chicks submitted to fasting or that received only water.

Table 3: Effect of egg weight and treatment type and duration on Villi Height (VH, mm) and Number (VN), Crypt Depth (CD, mm) and Globet Cells Number (GCN) in ileum of male broiler chicks

	VH	VN	CD	GCN
Eggs Weight (EW)				
Light eggs	0.93±0.13 B	8.10±0.72	0.06±0.01 B	15.42±3.09 B
Heavy eggs	0.99±0.10 A	8.38±1.00	0.07±0.01 A	18.81±2.90 A
Treatment Types (TT)				
Water + ration	1.01±0.13 A	7.72±0.89 B	0.08±0.01 A	17.03±3.90 B
Water	0.91±0.12 B	8.72±0.73 A	0.07±0.01 B	18.73±3.08 A
Fasting	0.96 ±0.09 B	8.39±0.75 A	0.07±0.01 B	16.23±2.70 B
Treatment Duration (TD)				
24 h	0.91±0.11 B	8.34±0.67 A	0.07±0.01	16.35±2.54 B
48 h	0.99±0.11 A	8.18±0.78 AB	0.07±0.01	18.14±3.33 A
72 h	1.00±0.13 A	7.76±0.87 B	0.07±0.01	18.00±3.95 A
Probability				
EW	0.0350	0.1120	0.0196	0.0001
TT	0.0004	0.0001	0.0012	0.0001
TD	0.0024	0.0001	0.8717	0.0001
EW x TT	0.2171	0.2219	0.0013	0.0003
EW x TD	0.6928	0.6771	0.5411	0.3406
TT x TD	0.0024	0.1257	0.4396	0.0022

A-B: Means followed by different letters (columns) are significantly different ($p \leq 0.05$)

The significant effect ($p \leq 0.05$) of the treatment duration on Jejunal Crypt Depth (JCD) and the significant interaction ($p \leq 0.05$) between egg weight and treatment type are presented at Table 2. The JCD was higher at 72 h than at 48 h (Table 2). Neither the JCD of chicks from light eggs nor the JCD of chicks from heavy eggs was affected by the treatments. However, differences in the JCD between chicks from light and heavy eggs were observed when they were fed with water and ration (Table 5).

The Globet Cells Number (GCN) in jejunum was influenced ($p \leq 0.05$) by egg weight and type and duration of the treatments (Table 2). This parameter was higher in chicks from heavy than those from light eggs. Chicks with access to water and ration or only to water had higher GCN than chicks submitted to fasting of feed and water. The GCN increased gradually during the experimental period (from 24-72 h).

Ileum: There was significant effect ($p \leq 0.05$) of the egg weight, type and duration of treatments for Ileal Villi Height (IVH), as well as significant interaction ($p \leq 0.05$) between treatment type and duration (Table 3). The villi were higher in chicks from heavy than from light eggs and in fed chicks than in chicks submitted to fasting of feed and water and only feed, which presented similar villi height. The IVH increased from 24-48 h and did not change between 48-72 h. The analysis of the interaction (Table 4) shows that the difference in the IVH among treatments occurred only at 72 h. The treatments did not differ at 24 and 48 h. The interaction also showed that the increase in IVH from 24-48 h occurred in chicks with access to water and ration and only to water. The fasted chicks presented the same IVH values during the experimental period.

There was significant effect ($p \leq 0.05$) of the treatment type and duration on Ileal Villi Number (IVN), but did not occur effect of the egg weight and interactions (Table 3). Fed chicks presented lower IVN than fasted chicks and chicks with access only to water. The IVN was lower at 72 than at 24 h.

Significant effect ($p \leq 0.05$) of egg weight and treatment type, as well as significant interaction ($p \leq 0.05$) between these variables on Ileal Crypt Depth (ICD) were detected (Table 3). The crypt was deeper in chicks from heavy than in light eggs and in fed than in fasted chicks and chicks with access to water, that did not differ one another. However, the interaction (Table 5) showed that differences in ICD between chicks from heavy and light eggs only occurred when they were fed. Besides, it was detected no differences in ICD among the treatments for chicks from light eggs. However, deeper crypts were observed in fed chicks from heavy eggs than those submitted to fasting of feed and water or with access only to water.

The Table 3 shows significant effect ($p \leq 0.05$) of the egg weight and treatment type and duration for GCN on ileum and significant interaction ($p \leq 0.05$) between type and duration of the treatments and between egg weight and treatment type. Chicks from heavy eggs presented higher GCN than chicks from light eggs. Chicks with access to water presented higher GCN than the fed and fasted chicks. The GCN increased from 24-48 h, remaining unchanged from 48-72 h. The first interaction (Table 4) showed difference in GCN between treatments only at 48 h, when chicks with access to water presented higher GCN than the fed and fasted chicks. Fed chicks and those with access only to water presented increase in GCN from 24-72 h. According to the interaction between egg weight and treatment type (Table 5), chicks from heavy eggs presented higher GCN than chicks from light eggs when they were fed or submitted to feed and water withdrawal. The GCN between chicks from heavy and light eggs presented no differences when they had access only to water.

Table 4: Interactions between type and duration of the treatments for Villi Height (VH, mm) in duodenum, jejunum and ileum; for Crypt Depth (CP, mm) in duodenum and for Globet Cells Number (GCN) in duodenum and jejunum of male broiler chicks

		24 h	48 h	72 h
Duodenum				
VH	Water + ration	1.72±0.20 Ab	1.94±0.30 Aab	2.08±0.32 Aa
	Water	1.60±0.18 Ab	1.85±0.23 Aba	1.86±0.23 Aa
	Fasting	1.68±0.17 Aa	1.55±0.28 Ba	1.56±0.22 Ba
CD	Water + ration	0.08±0.01 Aa	0.07±0.01 Aa	0.07±0.01 Aa
	Water	0.07±0.01 Aa	0.07±0.00 Aa	0.07±0.01 Aa
	Fasting	0.08±0.01 Aa	0.07±0.00 Ab	0.06±0.00 Ac
GCN	Water + ration	14.38±1.68 Ab	15.41±2.35 Aab	19.17±2.95 Aa
	Water	15.97±3.16 Aa	16.71±2.48 Aa	16.71±2.78 Aa
	Fasting	13.44±3.15 Ab	14.89±3.29 Ab	18.02±3.58 Aa
Jejunum				
VH	Water + ration	0.86±0.15 Bb	1.15±0.15 Aa	1.21±0.16 Aa
	Water	0.91±0.17 ABb	1.08±0.08 Aa	1.07±0.14 Aa
	Fasting	1.04±0.12 Aa	1.03±0.27 Aa	1.00±0.18 Ba
Ileum				
VH	Water + ration	0.90±0.12Ab	1.04±0.10Aa	1.09±0.07Aa
	Water	0.86±0.11Aa	0.98±0.13Aa	0.90±0.12 Ba
	Fasting	0.96±0.06 Aa	0.94±0.11Aa	0.98±0.09Ba
GCN	Water + ration	15.44±1.73 Ab	17.10±3.18 Bb	18.44±2.98 Aa
	Water	15.98±2.78 Ab	20.87±2.41 Aa	19.62±3.22 Aa
	Fasting	16.13±1.15 Aa	16.55±3.16 Ba	16.04±2.57 Aa

A-B, a-b: means followed by different letters (columns and lines, respectively) are significantly different ($p \leq 0.05$)

Table 5: Effect of the interactions between egg weight and treatment type on Crypt Depth (CD, mm) in duodenum, jejunum and ileum; on Villi Number (VN) in duodenum and jejunum and on Globet Cells Number (GCN) in ileum of male broiler chicks

		Water + ration	Water	Fasting
Duodenum				
CD	Light eggs	0.07±0.01 Bb	0.07±0.00 Bb	0.08±0.01 Aa
	Heavy eggs	0.09±0.01 Aa	0.08±0.01 Ab	0.07±0.01 Ab
VN	Light eggs	6.79±0.59 Ab	7.84±0.59 Aa	7.45±0.71 Aa
	Heavy eggs	7.23±0.97 Aa	7.23±0.97 Aa	7.03±0.56 Aa
Jejunum				
CD	Light eggs	0.07±0.01 Ba	0.07±0.01 Aa	0.07±0.01 Aa
	Heavy eggs	0.08±0.01 Aa	0.07±0.01 Aa	0.07±0.01 Aa
VN	Light eggs	6.93±0.45 Bb	8.44±0.58 Aa	8.60±0.94 Aa
	Heavy eggs	7.50±0.98 Ab	8.50±0.83 Aa	8.04±1.06 Aa
Ileum				
CD	Light eggs	0.07±0.01 Ba	0.07±0.01 Aa	0.07±0.01 Aa
	Heavy eggs	0.08±0.01 Aa	0.07±0.01 Ab	0.07±0.00 Ab
GCN	Light eggs	14.32±1.16 Bb	18.17±4.02 Aa	14.01±1.67 Bb
	Heavy eggs	19.40±3.91 Aa	19.14±2.22 Aa	17.83±2.10 Aa

A-B, a-b: Means followed by different letters (columns and lines, respectively) are significantly different ($p \leq 0.05$)

DISCUSSION

The height and number of villi are important parameters for analysis of villi growth, providing informations about the dynamic in digestive and absorptive intestinal areas. Crypt depth is determined by mitoses from stem cells present at the crypt base and is related with the dynamic of villi size regulation.

According to the results found in this experiment, fed chicks from heavy eggs when compared to fed chicks from light eggs presented higher villi and the same villi number in duodenum and ileum and higher villi and villi number in jejunum. These results indicate that fed chicks from heavy eggs have greater digestive and absorptive intestinal area than fed chicks from light eggs, explaining the better body weight of the first when compared to the second, as reported by Gimenez *et al.* (2008) and Riccardi *et al.* (2009). When fed, both chicks from light and heavy eggs presented during the 72 h of age an increase in villi height and decrease in villi number in duodenum, jejunum and ileum, simultaneously to the maintenance of the crypt depth. Villi size is determined by disequilibrium between mitosis rate of the crypts and cell extrusion rate at the apical extremity of the villi (Maiorka *et al.*, 2002). The villi growth occurs when mitosis rate becomes higher than the extrusion rate or when the extrusion rate decreases in relation to mitosis rate. Thus, the simultaneous villi growth and maintenance of crypt depth reported in the present experiment suggest the occurrence of low extrusion rate in relation to mitosis rate in the small intestine of fed chicks. The greater development of intestinal villi of chicks from heavy eggs during the first days posthatch should be also related to the high villi size, as reported by Gimenez *et al.* (2008).

The effects of delayed access to feed on DVH, JVH and IVH of chicks from heavy eggs did not differ from those

observed to chicks from light eggs, showing that the fasting influenced the villi height of both chicks from heavy and light eggs in a similar form and degree. The results found in this experiment also show that the fasting of feed and water inhibited the villi growth in duodenum, jejunum and ileum during the experimental period, but this effect did not begin simultaneously in all intestinal segments. In duodenum, the negative effect of the fasting on villi height occurred from 48 h, while in jejunum and ileum this effect occurred after 72 h. These results are in agreement with those obtained by Gomide *et al*. (2004), that also showed higher sensitivity of duodenum than jejunum and ileum to posthatch fasting. Considering that chicks use the nutrients from yolk sac in the first days posthatch (Vieira and Moran, 1999), the low negative effect of the fasting on jejunum and ileum can be related to the absorption of yolk sac nutrients by these cited intestinal segments. The present experiment also shows that water intake avoided the negative effects of fasting on DVH and JVH.

The Villi Number (VN) showed that the fasting of feed and water increased the intestinal VN, indicating absence of villi growth. However, chicks from light eggs presented this effect in duodenum, jejunum and ileum, while chicks from heavy ones presented only in jejunum and ileum. Besides, the increase in JVN and IVN was higher in chicks from light (9% and 20%, respectively) than from heavy eggs (0% and 7%, respectively). These results show that the posthatch fasting made impossible the growth of villi width and that the effect of the fasting was higher on chicks from light than from heavy eggs. The increase of the intestinal VN in chicks submitted to fasting was also reported by Maiorka *et al*. (2002, 2003). However, the results of this present experiment showed differences in intestinal mucosa response to fasting between chicks from light and heavy eggs, revealing a higher sensitivity of the chicks from light eggs. The results also demonstrated that the water intake did not avoid the fasting effects on intestinal VN of chicks from light and heavy eggs.

According to the results found in this present experiment, the fasted chicks from light and heavy eggs did not differ for DCD, JCD and ICD. Besides, during the fasting of feed and water the chicks presented reduction in DCD, while the JCD and ICD did not change. These results indicate a interruption of the mitosis in crypts, explaining the absence of villi growth observed in this present experiment for the small intestine of fasted chicks. The results also showed that the difference in CD reported between fed chicks from light and heavy eggs was kept by water intake only in duodenum. This experiment also showed that fasted chicks presented gradual reduction in DCD from 24-72 h that was not observed for chicks with access to water and ration or only water. Significant reductions in villi height and crypt depth of fasted chicks were also observed by Uni *et al*. (1998) and Geyra *et al*. (2001). However, different from these authors, this present experiment detected the effect of fasting on all intestinal segments, while they analyzed the effects only on duodenum and jejunum.

The Globet Cells Number (GCN) is a indicator of mucus production, which acts as protection barrier against chemical and microbiological damages and influences the nutrient absorption (Forstner and Forstner, 1994). For all treatments, the chicks from heavy eggs presented higher GCN in duodenum and jejunum than chicks from light eggs. However, in ileum this difference was observed only when the chicks did not have access to ration and water. Gimenez *et al*. (2008) verified that chicks from heavy eggs had higher villi than those from light eggs, indicating a greater digestive and absorptive area of chicks from heavy eggs and consequently the higher necessity of more GC to produce mucus and to protect the intestinal epithelium.

Data showed that the fasting of feed and water did not influence the GCN in duodenum and ileum, but decreased these cells in jejunum of chicks from heavy and light eggs. These results are in agreement with those found by Geyra *et al*. (2001) who observed the reduction of GCN after fasting. However, these results differed from those described by other authors, who observed reduction in GCN of ileum. The chicks after hatching present yolk sac that is weigher in chicks from heavy eggs than from light ones, having or not access to ration and water (Riccardi *et al*., 2009). During the experimental period in this present study, both fasted and fed chicks presented yolk sac from which they could obtain nutrients for maintenance and growth. Considering the yolk sac is linked to jejunum that is the local of absorption of the youlk sac nutrients (Peebles *et al*., 1998), it is possible that the reduction in jejunal GCN occurred in this experiment because of a lower necessity of protection against the fasting in this segment. The maintenance of the GCN in duodenum and ileum can be related to mucosa protection against gastric acids and enzymes action, as well as against the burning action of the yolk sac content. Besides, the water intake maintained the GCN in jejunum compared to water and ration supply in chicks from light and heavy eggs, but increased the GCN in ileum of chicks from light eggs. These results are very interesting because indicate the increase in mucus protection of ileum as response to liquid diet in chicks from light eggs.

Fed chicks from heavy and light eggs presented high GCN from 24-72 h in all intestinal segments simultaneously to villi growth, indicating that the mucus production increases proportionally with the growth of the interne intestinal surface. The increase of GCN in small intestine of chicks during the first week of age was also reported by Uni *et al*. (2001). The results referring to globet cells also show that fasted chicks did not present increase in iliacal GCN during the experimental period and this fact can be explained in function of the absence of villi growth in this intestinal segment. Besides, it can be observed that the water intake avoided the effect of the fasting on ileum and promoted increase of duodenal

GCN, in spite of the absence of villi growth in height. These results evidenced differences in the response to fasting among the different intestinal segments. This fact appears to be contradictory when considering that the mucus produced by the globet cells acts protecting the intestinal epithelium. However, it is possible that the maintenance of GCN when the chicks had only water supply is resulted from the less intensive effect of the gastric juice and enzymes on duodenal mucosa.

In conclusion, chicks from light eggs show major sensitivity to posthatch fasting than chicks from heavy eggs in many parameters. Besides, the water intake avoids partially the effects of the fasting on the analyzed parameters.

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