

*Research Article***Development of small intestinal morphology on the basis of growth and absorption rate in Broiler chicken (Cobb 500) of Bangladesh****Sohel, M. S. H.* , Miah, M. H., Faruq, A. A., Rahman M. L. and Islam, K. N.**

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Broiler, duodenum, jejunum, ileum, goblet cell, villi.

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E-mail: s.h.sohel08@gmail.com**ABSTRACT**

A gross and histomorphometric study was undertaken to evaluate the development of small intestinal morphology on the basis of growth and absorption rate in broiler chicken (Cobb 500) in Chattogram, Bangladesh from June to July, 2017. A total of 60 different ages (day 1, day 7, day 14, day 21 and day 28) broiler chickens were collected from a standard local broiler farm in Chittagong, Bangladesh. The live weight, gross length and diameter, villi length and diameter, thickness of tunica muscularis and the number of goblet cells of small intestinal segments (duodenum, jejunum and ileum) were studied. The tissue samples were fixed, processed and stained with Hematoxylin and Eosin stain and image measurement software was used for histomorphometric study. The average length and diameter of duodenum, jejunum and ileum were significantly higher in chicken at day 28 than that of others (day 21, day 14, day 7 and day 1). The villi of the duodenum, jejunum and ileum were lined by simple columnar epithelium. The length, width and goblet cells of villi and thickness of tunica muscularis of the duodenum, jejunum and ileum of broiler chicken were increased with the age significantly ($P \leq 0.05$). The highest mean value of villi length ($282.39 \pm 4.90 \mu\text{m}$), width ($38.49 \pm 9.68 \mu\text{m}$), goblet cells (1291.20 ± 26.10 per mm^2) and thickness of tunica muscularis ($75.34 \pm 1.67 \mu\text{m}$) were found in jejunum at 28 days old broiler chickens. These results indicate that the highest growth and absorption of broiler chicken (Cobb 500) occur in the jejunum at 21 to 28 days. So, it is suggested that the longer and wider villi as well as large number of goblet cells increase the growth and absorption rate of broiler chicken (Cobb 500).

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1. INTRODUCTION

The intestine is a very long tube which starts from the pyloric end of the stomach and terminates at the cloaca. Most of its parts are situated in the abdominal cavity.

Broadly, it is divided into two parts such as small intestine and large intestine. Small intestine starts from pylorus and terminates at the ileocecal junction (Getty 1975). The length and weight of the small intestine

fluctuated among different species of birds (Hassouna 2001). It is well known that small intestine is not only the most important digestive organ and the main absorptive site of the gastrointestinal tract, but also an immunological gut barrier to protect against invasion (translocation) of endogenous luminal microorganisms and/or their toxins (Ziegler *et al.*, 2003). The growth rate of broilers is related to intestinal development (Smith *et al.*, 1990). The weight of gastrointestinal tract of broiler chicken is about 1.5% of body weight. The relative growth of small intestine of broiler reaches its highest at 6 to 10 days of age in the presence or absence of feed (Mateos *et al.*, 2004; Sklan 2001).

Crypts and villi of the absorptive epithelium of small intestine play vital roles in the last stage of nutrient digestion and absorption. The absorption capacity of birds is changing due to the differential development of the absorptive epithelium (Verdal *et al.*, 2010). Studies on the small intestine have published that the size of the small intestine and its digestive activities are altered during development. In general, to understand the capacity of the small intestine to absorb nutrients, it is important to examine the morphological changes occurring during development. To date, very few studies were conducted on the development of small intestine of broiler chicken (Cobb 500) at different ages. There did not find out the morphological structures at different ages of bird supplement by different diets. Thus, the present study was conducted to know the gross and histological structure of small intestine and the correlation between age and the absorbable surface of the small intestine of broiler.

2. MATERIALS AND METHODS

A total 60 broiler chickens, 12 for each group of 1 day, 7 days, 14 days, 21 days and 28 days were collected from a standard local broiler farm in Chattogram, Bangladesh where feed and water has been supplied according to Banerjee (1978). At first, the chickens were sacrificed by excess chloroform inhalation, then the abdomen was cut, open and the entire small intestine was removed. The gross length and diameter of the duodenum, jejunum and ileum were measured by using a calibrated scale and recorded in centimeter (cm). Tissue samples (approximately 5–6mm) were obtained from the midpoints of the three segments. Then the tissue samples were fixed by using 10% neutral buffered formalin solution for a period of 72 hours. Subsequently the samples were processed through ascending and descending grade of alcohol and xylene, embedding in paraffin and sectioning into 5–6µ

m thickness by rotatory sliding microtome machine (LEICA SM2010RV1.2 English-09/2008 Sliding Microtome Machine, Germany) with the help of a disposable carbon blade. Both vertical and transverse sections were taken from each sample. Finally the tissue samples were stained following standard Hematoxylin and Eosin staining method (Gridley 1960). The villus length, width and diameter of muscular layer of duodenum, jejunum and ileum were measured by using a photograph microscope (AmScope Trinocular compound microscope and 1.3 MP camera, Model T490 B-MT) and Top view 3.0 image processing software. The densities of goblet cells were studied of vertical sections using transparent sheet on the slide whose one square is 1mm². The measurements were taken under a light microscope (Olympus, CH2XX, Tokyo, Japan) at 10×objective. Then all data were entered into a Microsoft excel sheet-2007 and transferred to STATA-13 to perform. If the p-values were equal or less than 0.05 ($P \leq 0.05$) then the differences were considered as significant. The data were expressed as mean \pm SE.

3. RESULTS AND DISCUSSION

Gross morphometry of duodenum, jejunum and ileum

Table 1 showing that the lengths and diameters of duodenum, jejunum and ileum were increased gradually with increasing age, where the lowest lengths (7.83 ± 1.27 cm, 22.75 ± 1.48 cm and 7.83 ± 1.59 cm, respectively) and diameters (0.18 ± 0.08 cm, 0.08 ± 2.84 cm and 0.19 ± 0.08 cm, respectively) were found at day 1 and the highest lengths (36.00 ± 1.41 cm, 118.25 ± 4.88 cm and 31.33 ± 2.10 cm, respectively) and diameters (1.31 ± 0.14 cm, 1.54 ± 0.17 cm and 1.02 ± 0.09 cm, respectively) were found at 28 days old broiler chicken. These results are similar to Hassouna (2001); Nasrin *et al.*, (2012) but Getty (1975) reported that the lengths of duodenum, jejunum and ileum of chicken were 22–35cm, 85–120cm, 13–18cm, respectively and the diameters were 0.18–1.31cm, 0.08–1.54cm, 0.19–1.02cm, respectively. These figures also show that the growth rate of jejunal lengths and diameters was very fast. On the other hand the lengths and diameters of ileum increased faster than duodenum but slower than jejunum.

Histomorphometry of duodenum, jejunum and ileum

Histomorphometric study shows wide variation in the structure of the intestinal wall, particularly the absorptive epithelium (villi) and the muscular layer. The villi of the duodenum, jejunum and ileum of

Tables

Table 1. Gross morphometry of duodenum, jejunum and ileum. Measures are given as Mean \pm Standard error, N = 60.

Age (Day)	Duodenum		Jejunum		Ileum	
	Length (cm)	Diameter (cm)	Length (cm)	Diameter (cm)	Length (cm)	Diameter (cm)
1	7.83 \pm 1.27	0.18 \pm 0.08	22.75 \pm 1.48	0.08 \pm 2.84	7.83 \pm 1.59	0.19 \pm 0.08
7	16.25 \pm 2.70	0.43 \pm 0.12	47.33 \pm 6.09	0.55 \pm 0.10	14.92 \pm 2.15	0.43 \pm 0.12
14	25.33 \pm 1.87	0.85 \pm 0.12	69.83 \pm 3.88	0.88 \pm 0.08	21.58 \pm 2.35	0.63 \pm 0.09
21	31.17 \pm 1.64	1.02 \pm 0.11	89.42 \pm 7.63	1.25 \pm 0.12	27.83 \pm 1.47	0.81 \pm 0.11
28	36.00 \pm 1.41	1.31 \pm 0.14	118.25 \pm 4.88	1.54 \pm 0.17	31.33 \pm 2.10	1.02 \pm 0.09

Table 2. Histomorphometry of duodenum, jejunum and ileum. Measures are given as Mean \pm Standard error, N = 75.

Age (Day)	Duodenum			Jejunum			Ileum		
	Villi		TM	Villi		TM	Villi		TM
	Length (cm)	Width (cm)	Diameter (cm)	Length (cm)	Width (cm)	Diameter (cm)	Length (cm)	Width (cm)	Diameter (cm)
1	100.3 \pm 6.69	18.02 \pm 3.3	20.44 \pm 2.92	132.97 \pm 9.21	24.34 \pm 2.99	22.58 \pm 2.55	140.59 \pm 6.32	18.79 \pm 3.54	21.23 \pm 4.75
7	127.6 \pm 2.47	18.79 \pm 3.5	21.00 \pm 5.08	168.24 \pm 9.64	24.91 \pm 6.73	31.94 \pm 5.10	191.44 \pm 10.1	20.54 \pm 3.76	26.62 \pm 6.52
14	128.7 \pm 6.70	20.45 \pm 3.8	27.70 \pm 1.85	169.80 \pm 7.90	25.34 \pm 2.11	41.14 \pm 5.02	168.86 \pm 7.95	21.44 \pm 2.12	28.14 \pm 5.23
21	146.5 \pm 2.30	20.52 \pm 7.1	30.46 \pm 3.15	191.58 \pm 3.80	28.92 \pm 7.30	41.17 \pm 1.77	212.68 \pm 5.56	28.60 \pm 6.90	30.52 \pm 3.06
28	143.6 \pm 1.29	19.15 \pm 9.5	30.53 \pm 5.45	282.39 \pm 4.90	38.49 \pm 9.68	75.34 \pm 1.67	274.19 \pm 8.74	35.50 \pm 7.87	30.77 \pm 5.68

TM = Tunica muscularis

Table 3. Goblet cells of villi of duodenum, jejunum and ileum. Measures are given as Mean \pm Standard error, N = 75.

Age (Day)	Goblet cells of villi (cells / mm ²)		
	Duodenum	Jejunum	Ileum
1	303.45 \pm 20.23	646.38 \pm 28.21	528.21 \pm 27.35
7	646.38 \pm 35.26	979.49 \pm 30.26	837.77 \pm 35.49
14	789.67 \pm 38.78	1024.48 \pm 30.12	906.40 \pm 35.20
21	987.57 \pm 20.35	1138.23 \pm 31.10	1120.27 \pm 41.29
28	1028.79 \pm 40.30	1291.20 \pm 26.10	1204.47 \pm 39.47

broiler chicken were lined by simple columnar epithelium (Figure 1-3). These analogous results were reported by Aitken (1958); Getty (1975); Nasrin *et al.*, (2012). The apical parts of villi of the duodenum and jejunum were slightly pointed and the basal parts were wider, but most of the villi of jejunum was blunted apical part and wider basal part as observed by Aitken (1958); Getty (1975) in chicken. Table 2 shows that the lengths and widths of villi of the duodenum, jejunum and ileum of broiler chicken were increased gradually with increasing age significantly ($P \leq 0.05$). The highest mean villus length ($282.39 \pm 4.90 \mu\text{m}$) and width ($38.49 \pm 9.68 \mu\text{m}$) was found in jejunum at 28 days old broiler chicken. These results completely agree with some previous observations (Hassouna 2001; Verdal *et al.*, 2010; Nasrin *et al.*, 2012). The growth rate of jejunum was much higher than the duodenum and ileum. This finding agreed with Hassouna (2001) but Verdal *et al.*, (2010) reported that the growth rate of ileum was much higher than the duodenum and jejunum of broiler chicken. From the histological point of view, it could have been expected that longer villi in the present study in broiler chicken (Cobb 500) resulted in an increased surface area that allowed greater absorption of available nutrients, particularly in the jejunum at 28 days old broilers

(Caspary 1992). Longer villi might consequently promote growth (Yamauchi *et al.*, 2006; Markovic *et al.*, 2009). Our study reveals that overall length and width of villi of jejunum was maximum at 28 days old broiler chicken. Thus we concluded that absorption rate was highest in jejunum at 28 days old broiler chicken.

In the present study, diameter of tunica muscularis of the duodenum, jejunum and ileum of broiler chicken were increased gradually with increasing age significantly ($P \leq 0.05$) (Table 2). The highest diameter ($75.34 \pm 1.67 \mu\text{m}$) was found in jejunum at 28 days old broiler chicken. These results are similar to Hassouna (2001); Verdal *et al.* (2010). The growth rate of tunica muscularis of the jejunum was much higher than the duodenum and ileum.

Table 3 represents that the number of goblet cells (per mm^2) in the villi of the duodenum, jejunum and ileum were significantly ($P \leq 0.05$) increased with increasing age. The highest number of goblet cells ($1291.20 \pm 26.10 \text{ cells/mm}^2$) was found in the jejunum at 28 days old broiler chicken than the others. These findings support the previous studies (Uni *et al.*, 2003; Geyra *et al.*, 2001). An overall finding of goblet cells of small intestine reveals that the absorption rate was maximum in jejunum at 28 days old broiler chicken.

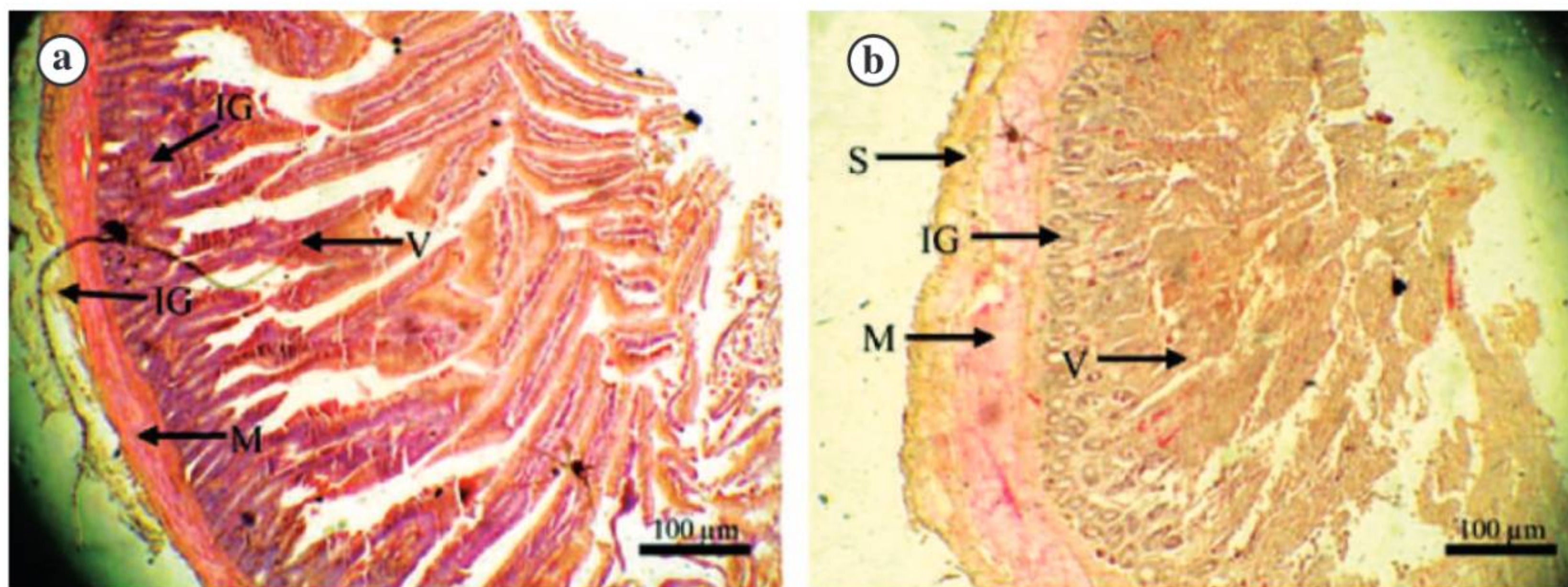


Figure 1. Histological structure of 14 days (a) and 28 days (b) old duodenum in H&E staining showing Villi (V), Intestinal gland (IG), Tunica muscularis (M), Serosa (S); Scale bar 100 μm .

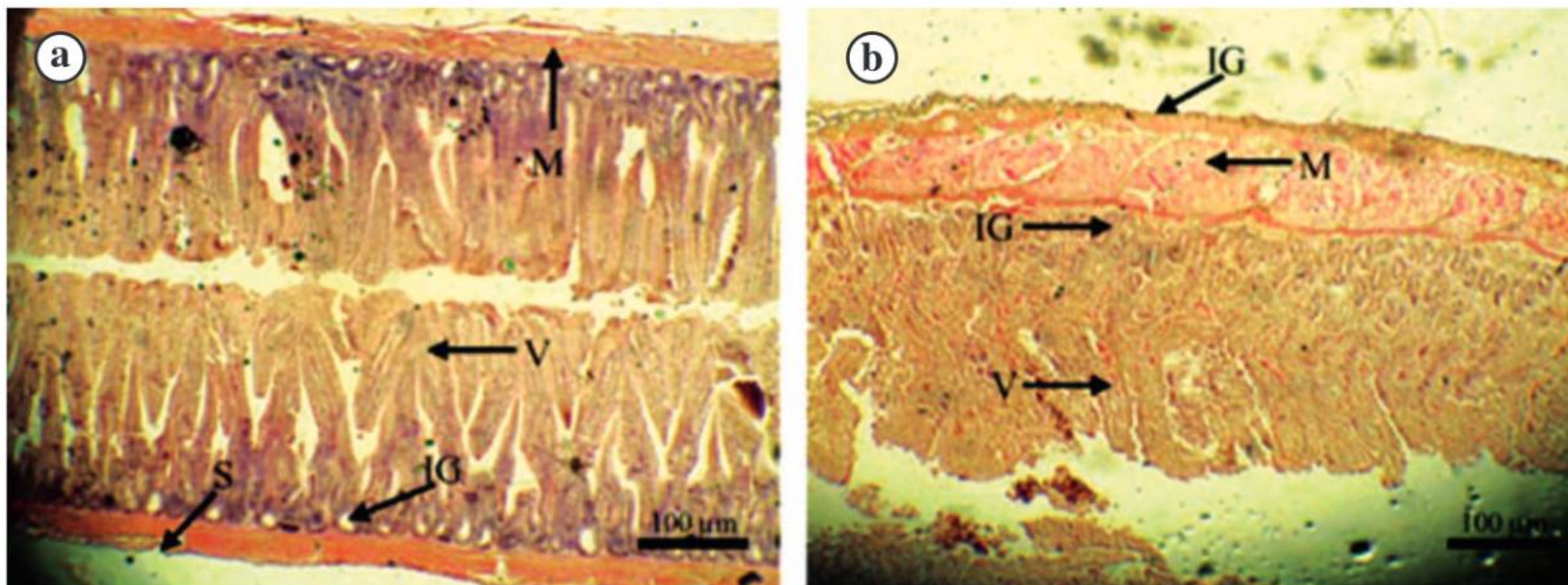


Figure 2. Histological structure of 14 days (a) and 28 days (b) old jejunum in H&E staining showing Villi (V), Intestinal gland (IG), Tunica muscularis (M), Serosa (S); Scale bar 100 μ m.

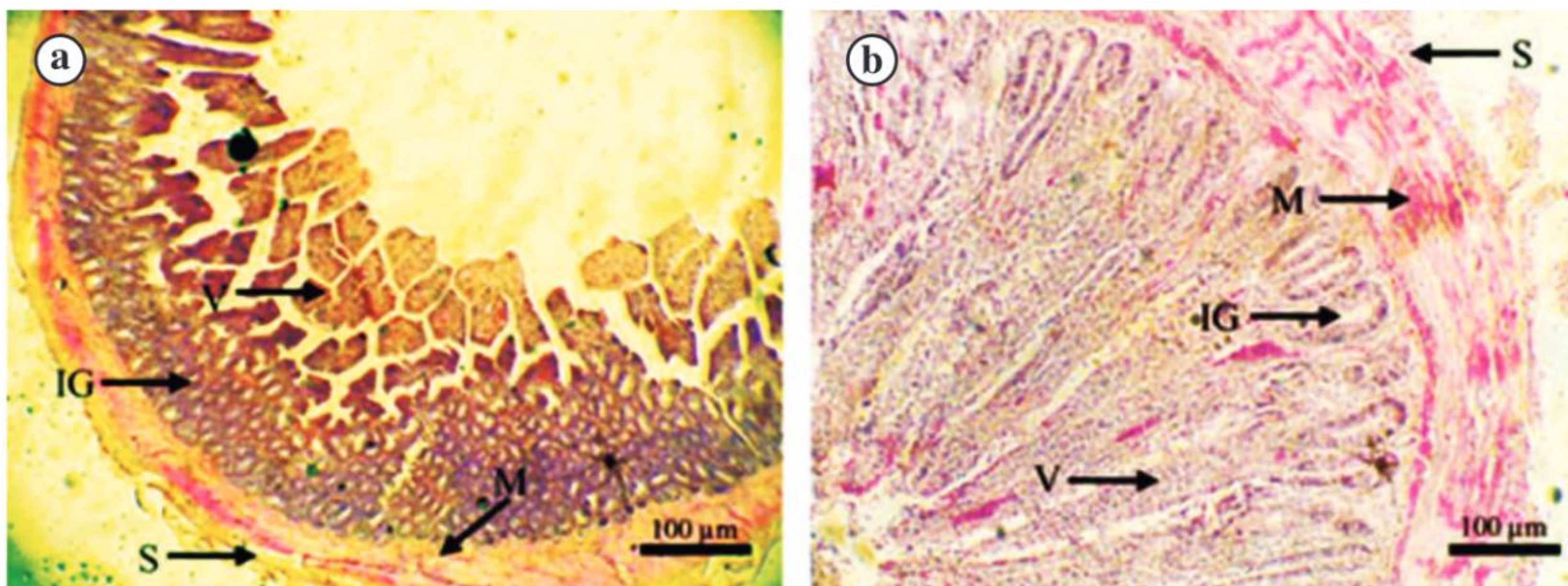


Figure 3. Histological structure of 14 days (a) and 28 days (b) old jejunum in H&E staining showing Villi (V), Intestinal gland (IG), Tunica muscularis (M), Serosa (S); Scale bar 100 μ m.

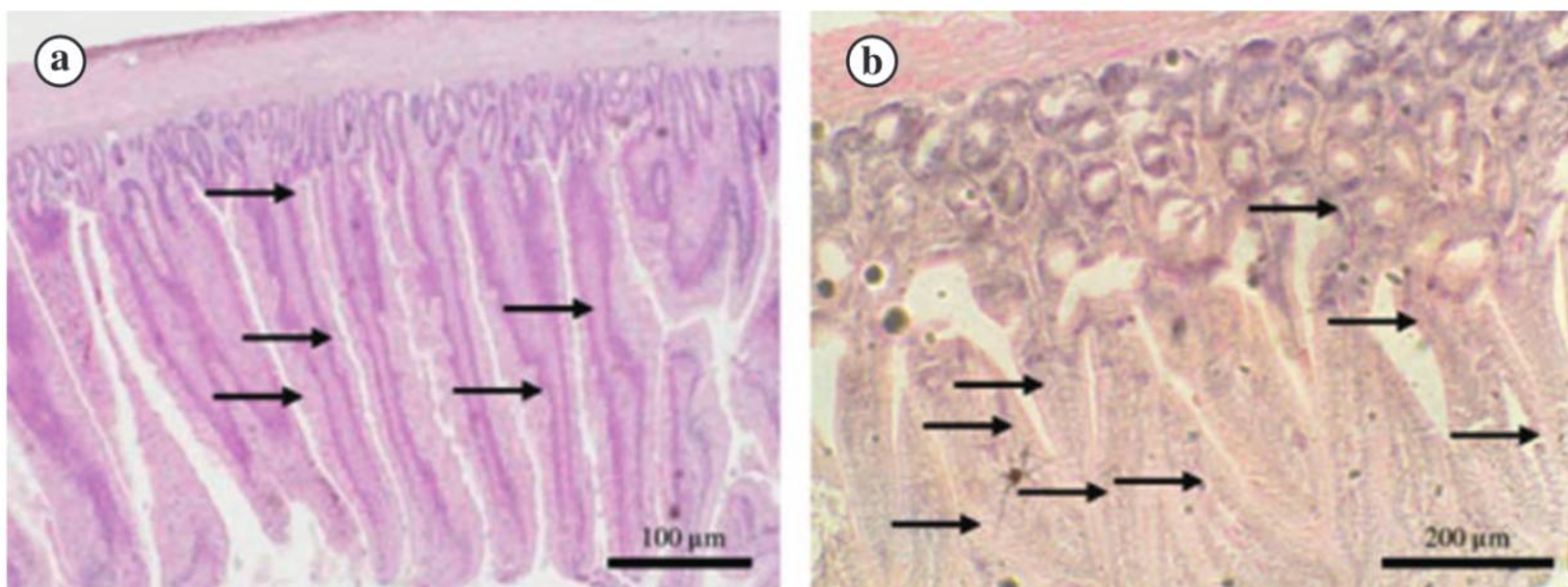


Figure 4. Histological structure of duodenum (a) and jejunum (b) in H&E staining where arrows showing goblet cells.

4. CONCLUSIONS

It is recommended that the growth and the absorption rate of broiler chicken (Cobb 500) varies with different age and it depends on the histomorphological characteristics of villi including length, width and the number of goblet cells of the villi of the small intestine.

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