Development of small intestine and sugar absorptive capacity in goslings during pre- and post-hatching periods

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ABSTRACT This study was conducted to investigate the development patterns of small intestine, intestinal morphology, disaccharidase activities, and sugar transporter gene expression in goslings during pre- and post-hatching periods. Small intestine was sampled on embryonic d 23 and 27, day of hatch, and d 1, 4, and 7 post-hatching. A total of 18 eggs with the breed of Jilin White geese were selected at each sampling timepoint for measuring relevant parameters. Three eggs were considered as a group, with 6 groups in each sampling timepoint. Rapid development of small intestine was observed around the hatching, of which jejunum and ileum had relatively higher development rates. Villus surface area from three intestinal segments started to increase on embryonic d 27, and kept relatively stable during day of hatch to d 1 post-hatching, and following increased till d 7 post-hatching. A high priority of villi enrichment was observed in duodenum and jejunum. The activity of disaccharidase increased before hatching and kept relatively high-level post-hatching, of which the activity of disaccharidase was highest in jejunum. The expression of sugar transporter gene increased prior to hatching and then decreased post-hatching, of which jejunum and duodenum were sites with high sugar transporter gene expression. Rapid development in intestinal morphology, disaccharidase activities, and sugar transporter gene expression around the hatching indicated that goslings have high potential to digest and/or assimilate carbohydrates during its early-life, which provided a preparation for further digestion of exogenous feed. This study provided a profile of development patterns for intestinal morphology, disaccharidase activities, and sugar transporter gene expression in goslings, which was beneficial to understanding the characteristics of nutrient absorption during the early-life of goslings.

Key words: gosling, small intestine, intestinal morphology, sugar transporter gene expression, disaccharidase

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INTRODUCTION

Geese are widely raised all over the world, which is a nutritious and healthy food resource. Because of the short reproductive periods, low hatchability, and high embryo mortality of geese, goose farming industry is not as prosperous as broilers industry (Tai et al., 2001; Rosinski et al., 2006a,b). However, it is reported that the content of protein and trace elements in the meat of goose is higher than in other poultry products (Xu et al., 2018). Therefore, goose husbandry has great economic prospects. Future goose farming husbandry will benefit from research on the organ development pattern of goslings. Among them, intestine plays a key supporting role in the growth of animals. It is very important to investigate the morphological development of small intestine, intestine internal disaccharidase activities, and sugar transporter gene expression during late-term period of incubation and the early period post-hatching to understand the nutrient requirements of goslings during their early-life.

Recently, a comprehensive investigation has been conducted on the development patterns of embryonic intestines of chicks, ducks, turkeys, guinea fowls, and pigeons (Dong et al., 2012a,b; Wilson et al., 2018; Araújo et al., 2019; Li et al., 2019; Givisiez et al., 2020). Dong et al.

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(2012a) observed the expression of intestinal nutrient transporters increased with age, moreover, they noted that the expression of sugar transporter genes in pigeon jejunum was higher than that in other intestinal segments. They further investigated the post-hatching development of intestinal morphology of pigeons and found a rapid development in intestinal morphology and digestive enzyme activities (Dong et al., 2012b). In turkey, Wilson et al. (2018) reported that the length and width of villi developed with age. Givisiez et al. (2020) reviewed the functional development of gastrointestinal tract in chicks, they noted that the morphological changes, digestive enzyme activities, and nutrient transporter gene expression developed with age. However, the embryonic development patterns in intestinal morphology, disaccharidase activities, sugar transporter gene expression of goslings during pre- and post-hatching periods have not been fully understood.

To understand the intestinal development patterns of small intestine and sugar absorptive capacity in goslings, we conducted this study to investigate the small intestine development pattern, intestinal morphology, and sugar transporter gene expression in goslings during pre- and post-hatching periods.

MATERIALS AND METHODS

This study was approved by the Animal Care and Use Committee of Jilin Agricultural University (Changchun, Jilin, China) with a code of S83520130804.

Experimental Animals

A total of 150 fertilized eggs (Jilin White Geese) used in this study were obtained from the Geese Experimental Central of Jilin Agricultural University. A commercial incubator (Keyu CFZ microcomputer automatic incubator, Dezhou, Shandong, China) was used to incubate eggs. Preheat the eggs (30℃ for 12 h) and sterilize them (37% formaldehyde and potassium permanganate in a ratio of 2:1), then move them into the incubator. Incubation period was divided into 3 phases: phase 1, embryonic d 1 to d 14, the temperature was 38℃ and the humidity was 65%; phase 2, embryonic d 15 to d 28, the temperature was 37.5℃ and the humidity was 55%; phase 3, embryonic d 29 to d 31, the temperature was 37.2℃ and the humidity was 70%. During the incubation period, eggs were turned every 2 h for 180 s.

On embryonic d 23, eggs were candled to remove unfertilized eggs. A total of 120 fertilized eggs with a similar weight were used for further studies. In this study, the sampling timepoints were embryonic d 23, d 27, day of hatch, and d 1, d 4, and d 7 post-hatching. At each sampling timepoint, a total of 18 eggs were randomly selected and randomly assigned to 6 groups with 3 replicates in each group.

After hatching, geese were transported to the farm and assigned to cages (25 birds per cage), and immediately received feed (Table 1). All geese were raised under uniform management conditions with 30°C. During the experimental period, birds had free access to receive feed and water.

Table 1. Composition and nutrient levels of the experimental basal diet, (%, as-fed basis).

Ingredients, %	
Corn	60.00
Soybean meal	29.11
Wheat bran	6.00
Fish meal	2.00
Lysine-HCl	0.20
Methionine	0.23
Dicalcium phosphate	0.84
Limestone	0.82
Sodium chloride	0.30
Vitamin and trace mineral premix[1]	0.50
Total	100.00
Calculated value, %	
Metabolizable energy, MJ/kg	11.67
Available phosphorus	0.40
Analyzed composition, %	
Calcium	0.78
Crude protein	19.78
Methionine	0.50
Total sulfate amino acid	0.77
Lysine	1.08
Crude fiber	0.31
Neutral detergent fiber	1.09
Acid detergent fiber	0.35

[1]Provided per kg of complete diet: vitamin D_3, 200 IU; vitamin A (retinyl acetate), 1,500 mg; vitamin E (DL-α-tocopheryl acetate), 12.5 mg; vitamin K_3, 1.5 mg; thiamine, 2.2 mg; riboflavin, 5 mg; nicotinic acid, 65 mg; folic acid, 1 mg; pantothenic acid, 15 mg; pyridoxine, 2 mg; biotin, 0.2 mg; choline, 1,000 mg; Fe, 90 mg; Cu, 6 mg; Mn, 85 mg; Zn, 85 mg; I, 0.42 mg; Se, 0.3 mg; Co, 2.5 mg.

Feed Analysis

Feed samples were dried using a thermostatic oven (70°C) for 72 h. Samples were then ground and sieved by a 1-mm sieve. Then, the contents of dry matter, crude protein, calcium, and crude fiber in the diet were analyzed according to methods provided by the Association of official analytical chemists (AOAC, 2000). Moreover, the contents of neutral detergent fiber and acid detergent fiber in the diet were analyzed according to methods provided by Mertens (2002). Before analyzing amino acid contents, feed samples were hydrolyzed with 6 N HCl at 110°C for 24 h. Then, the amino acid contents in the feed sample were analyzed by an amino acid analyzer (2690 Alliance, Waters, Inc., Milford, MA).

Sample Collection and Measurement

A pair of surgical scissors was used to open the eggs. Then, the embryo with yolk sac was weighed and the small intestine was taken out. Ice-cold saline was used to remove adherent materials and/or internal contents from the small intestine. Record the weight and length of small intestine, then divide it into duodenum, jejunum, and ileum. Intestinal segment samples were duplicate taken from the middle of intestinal segment with a length of 1-cm. Intestinal segments were stored individually in 2 tubes. One sample was fixed with 10% neutral-

Table 2. Primers used for quantitative real-time PCR.

Gene	Accession number[1]	Size (bp)[2]	Primer sequences (5'→3')	
β-Actin	M26111	158	Forward	GCCCAGCAC GATGAAGAT
			Reverse	ATTTACGGT GGACGATGGAC
SGLT1	KU744842	126	Forward	GTAACATTGG CAGCGGACAT
			Reverse	TGGGTACAAA CAGCCATCCT
GLUT2	KU744841	118	Forward	CAGTTCTTCC TGCTCCTGCT
			Reverse	TCATCGGGTC ACAGTTTCCT
SI	KU744844	193	Forward	CGTCACCTT CCCTCTTTGG
			Reverse	GGATTATGCTTC ACTTCCACTTTG

Abbreviations: GLUT-2, glucose transporter-2; SGLT-1, sodium/glucose cotransporter protein-1; SI, sucrase-isomaltase.
[1]Accession number refer to Genbank (NCBI).
[2]PCR product size (base pairs).

buffered formalin solution to measure histology, and the other sample was frozen in liquid nitrogen to measure the activities of disaccharidase and the expression of sugar transport genes.

Small Intestine Parameter Analysis

Below equation was used to measure the relative weight of small intestine:

$$Organ\ index\ =\ \frac{Organ\ weight}{Live\ body\ weight}\ \times\ 100\ \%.$$

A graduated ruler was used to measure the length of small intestine.

Small Intestine Morphology Analysis

Intestinal segment samples were cut into small pieces to measure the morphology according to the method described by Dang et al. (2022). In brief, small pieces of intestinal segment samples were fixed by 10% neutral buffered formalin for 12 h, and then dehydrated with alcohol of gradient concentration and xylene. Treated samples were then used to make paraffin blocks. A cryostat was used to make tissue sections. After removing paraffin, samples were then stained with hematoxylin and eosin. An optical microscope (Olympus, BX53F, Tokyo, Japan) was used to measure the values of villus height and width at 10X magnification. For each parameter, each slide was measured 5 times and represented on average. Villi area was calculated by the villi height (from the villi tip to the junction of villi crypt) and width at half height. Values given are averages from 10 adjacent villi, and only vertically oriented villi were measured.

Disaccharidase Activities Analysis

Intestinal segment samples were homogenized by 10 times of the volume of cold normal saline. The homogenates were then centrifuged at 3,500 × g at 4°C for 15 min for collecting supernatant. According to the method described by Dang et al. (2022), colorimetric method was used to measure the activities of sucrase and maltase.

Sugar Transport Gene Expression Analysis

Based on the method described by Dang et al. (2022), RNAiso Reagent (TaKaRa, Dalian, Liaoning, China) was used to isolate the total RNA from intestinal segment samples. The integrity and concentration of RNA were then determined. The primer sequences of the test gene were specially designed according to the sequences in GenBank (Table 2). Total RNA samples were purified by a specific kit, and then reverse transcribed, followed by cDNA synthesis. RT-PCR was used to analyze the relative expression levels of sodium/glucose cotransporter protein-1 (**SGLT-1**), glucose transporter-2 (**GLUT-2**), and sucrase-isomaltase (**SI**) mRNA isolated from geese intestinal segment tissues. β-actin was used as the internal reference.

Statistical Analysis

All data were analyzed using SPSS18.0 software. Tukey test was used for multiple comparisons among different ages. Values from each group (3 eggs) were pooled to form a sample. Variability in the data was expressed as the standard error of means (**SEM**). Results were considered significant at $P < 0.05$.

RESULTS AND DISCUSSION

The intestinal tract developed rapidly with the increase of body weight (Table 3). The weight and length of intestine, and its proportion to embryo weight are considered important parameters reflecting the development of the small intestine (Chen et al., 2021). The rapid development patterns of small intestine in the

Table 3. Body weight and small intestine parameters of goslings between embryonic d 23 to d 7 post-hatching[1].

Items	E23	E27	DOH	D1	D4	D7	SEM	*P*-value
Body weight, g	46.85[e]	69.33[d]	80.53[c]	80.29[c]	120.45[b]	158.96[a]	1.890	<0.01
Intestinal segments weight, g								
Duodenum	0.05[e]	0.14[e]	0.67[d]	0.82[c]	1.47[b]	2.01[a]	0.710	<0.01
Jejunum	0.15[f]	0.35[e]	1.11[d]	1.34[c]	3.07[b]	4.16[a]	1.483	<0.01
Ileum	0.15[e]	0.32[e]	1.15[d]	1.57[c]	3.14[b]	3.78[a]	1.379	<0.01
Intestinal segments length, cm								
Duodenum	5.00[d]	5.90[d]	9.33[c]	9.87[c]	13.50[b]	18.43[a]	4.749	<0.01
Jejunum	9.60[d]	11.37[d]	18.33[c]	19.67[c]	28.77[b]	37.10[a]	9.835	<0.01
Ileum	9.67[e]	10.37[e]	17.77[d]	19.83[c]	27.67[b]	34.83[a]	9.232	<0.01
Relative weight of intestinal segments, %								
Duodenum	1.07[d]	2.07[d]	8.48[c]	10.22[b]	12.28[a]	12.61[a]	4.754	<0.01
Jejunum	3.20[e]	5.00[d]	13.96[c]	16.65[b]	25.55[a]	26.14[a]	9.114	<0.01
Ileum	3.22[e]	4.56[e]	14.39[d]	19.66[c]	26.18[a]	23.76[b]	9.118	<0.01

[a-f]Different superscripts within a raw indicate a significant difference ($P < 0.05$).

[1]Age refers to embryonic d 23 (**E23**), d 27 (**E27**), day of hatch (**DOH**; after hatch but before feeding), and d 1 (**D1**), d 4 (**D4**), and d 7 (**D7**) post hatching.

late-term embryonic development have been confirmed in chicks and pigeons (Uni et al., 2003a,b; Dong et al., 2012a,b). Uni et al. (2003b) reported that the proportion of intestine to embryo weight increased rapidly in the last 3 days of incubation in chicks. Dong et al. (2012b) also observed that the intestine of pigeons developed rapidly in the late-term of incubation. In the present study, we also observed a rapid development of small intestine during the late-term of incubation (Table 3), of which the increase in duodenal and ileal weight, length, and those proportion to embryo weight occurred on embryonic d 27. Additionally, we observed that the weight of jejunum and its proportion to embryo weight increased continuously during this period. The rapid development in small intestine during the late-term of incubation provided a preparation to receive the exogenous feed after hatching. Subsequently, the development of small intestine continued at a high-speed during post-hatching (Table 3). The weight and length of these 3 intestinal segments and the proportion of intestinal segments to the embryo weight increased continuously during the post-hatching period. A similar development pattern in chicks was also reported by Katanbaf et al. (1988), they noted that the intestine developed with age. The rapid development of small intestine after hatching may be attributed to the stimulating effect of exogenous feed on small intestine. Jin et al. (1998) reported that the relative weight of the whole intestinal tract among the birds receiving exogenous feed in time increased by approximately 20% during the first 5 d post-hatching, however, there was almost no change in fasting chicks during this period. A good development of intestinal tract laid a physiological foundation for realizing the maximum growth potential, which was crucial for the growth of goslings. As the results observed in this study, the intestinal tract of goslings developed rapidly during the late-term of incubation and the early-life post-hatching, which contributed to supporting the increase of body weight. However, the development patterns in different intestinal segments were different. Dror et al. (1977) and Uni (1999) reported the highest growth rate of intestinal segments in chicks was duodenum, followed by jejunum and ileum. In contrast, this study observed that jejunum and ileum were the segments with relatively high growth rates, followed by duodenum. Additionally, we observed a reduction effect in the relative weight of ileum from d 4 post-hatching to d 7 post-hatching, which probably indicated that the high-speed development of ileum was stopped/retarded on d 4 post-hatching, however, this statement also needed to be verified by further studies. To sum up, we considered that jejunum and ileum have a high priority in intestinal growth of goslings during early-life. Unlike chicks, geese have a large body size and strong tolerance/adaptability to roughage (Li et al., 2017), which may determine its unique intestinal segment development patterns.

Moreover, the development of small intestine is not only presented in the improvement of the apparent parameters of small intestine, but also in the variation of its morphological structure. The height and width of villi, and its surface area are commonly used parameters to reflect the function of nutrient absorption (Rajput et al., 2012). The epithelium of small intestine protrudes into gut lumen to form long folds, that is, villi, which are the functional units of small intestine involved in nutrient absorption. In this study, we observed the increase of villus absorptive area in three intestinal segments started on embryonic d 27, after incubation, it kept relatively stable during day of hatch to d 1 post-hatching. Subsequently, on d 1 post-hatching, it continued to increase till d 7 post-hatching (Table 4). Similarly, Sklan (2001) and Uni (2006) reported that the morphology of the small intestine changed rapidly during the early-life of chicks. The optimization of villi morphology laid the physiological foundation for nutrient absorption. However, the villi have different ontogenetic timetables in different intestinal segments (Sklan, 2001). Sklan (2001) and Scanes and Pierzchala-Koziec (2014) detailed small intestinal morphological development of chicks and found duodenum was almost covered completely by villi on d 7 post-hatching, while not jejunum and ileum. Baranyiova and Holman (1976) investigated the development status of villi in chicks immediately after hatching,

Table 4. Small intestine morphology parameters of goslings between embryonic d 23 to d 7 post-hatching[1].

Items	E23	E27	DOH	D1	D4	D7	SEM	*P*-value
Villus height, μm								
Duodenum	55.88^{d}	80.09^{d}	202.12^{c}	206.14^{c}	247.08^{b}	321.68^{a}	98.545	<0.01
Jejunum	46.72^{d}	73.71^{d}	230.83^{c}	218.35^{c}	281.73^{b}	390.20^{a}	121.566	<0.01
Ileum	39.38^{c}	61.66^{c}	196.10^{b}	208.68^{b}	207.51^{b}	243.09^{a}	82.332	<0.01
Villus width, μm								
Duodenum	24.82^{d}	18.68^{d}	42.81^{c}	45.62^{c}	59.69^{b}	69.81^{a}	19.008	<0.01
Jejunum	26.41^{c}	17.63^{d}	33.04^{bc}	37.46^{b}	40.38^{b}	63.36^{a}	15.842	<0.01
Ileum	35.92^{b}	20.55^{d}	27.65^{c}	28.77^{c}	34.83^{b}	42.55^{a}	7.584	<0.01
Villus area, $\mu m^2 \times 10^3$								
Duodenum	1.38^{d}	1.50^{d}	8.58^{c}	9.42^{c}	14.47^{b}	22.29^{a}	7.600	<0.01
Jejunum	1.25^{d}	1.30^{d}	7.65^{c}	8.21^{c}	11.42^{b}	24.70^{a}	8.333	<0.01
Ileum	1.42^{d}	1.26^{d}	5.42^{c}	5.92^{c}	7.28^{b}	10.39^{a}	3.379	<0.01

[a-d]Different superscripts within a raw indicate a significant difference ($P < 0.05$).
[1]Age refers to embryonic d 23 (E23), d 27 (E27), day of hatch (DOH; after hatch but before feeding), and d 1 (D1), d 4 (D4), and d 7 (D7) post hatching.

they observed that the longest villi appeared in duodenum, and its length was twice that of jejunum and ileum. Additionally, Uni et al. (1998) and Uni (1999) reported that the development of duodenal villi in chicks was completed around d 6 or 7 post-hatching, whereas the development of jejunal and ileal villi will be completed till d 14 post-hatching. Therefore, results reported by the above studies seem to indicate that duodenal villi in chicks have a high priority in the development of intestinal villi. In contrast, in the present study, we observed that duodenum and jejunum had similar villi surface area, which was higher than those of ileum (Table 4). Therefore, in goslings, we considered that villi enrichment has a high priority in duodenum and jejunum, followed by ileum.

The activities of intestinal digestive enzymes increased with the development of intestinal morphology (Moosavinasab et al., 2015). In chicks, Uni et al. (2003a, b) and Uni (2006) reported that the total intestinal disaccharidase activities started to rise during the last 2 d before hatching and increased rapidly post-hatching. In pigeons, Dong et al. (2012a,b) noted that the activity of mucosal and total intestinal disaccharidase increased with age. Similarly, in this study, we observed that the sucrase activity in three intestinal segments started to increase on embryonic d 27 and continued to increase with age (Table 5). The activity of maltase in three intestinal segments started to increase on embryonic d 23 and increased gradually with age, but kept relatively stable on d 4 post-hatching (Table 5). Moreover, from d 4 post-hatching to d 7 post-hatching, we observed a decrease in duodenal maltase activity, which probably indicated that duodenum was not the primary site for maltose digestion (Uni et al., 1998). Indeed, the regional activity of mucosal enzymes was different in these three intestinal segments. In chicks, it was reported that jejunum has the strongest ability to digest disaccharides, followed by ileum and then duodenum (Uni et al., 1998). In the present study, we observed the highest disaccharidase activities occurred in jejunum, followed by ileum and then duodenum, which was the same as the distribution of disaccharidase activities in the intestinal segments of chicks (Uni et al., 1998). Therefore, we considered that jejunum was the main site for disaccharide digestion in goslings.

Digestion occurs when carbohydrates are degraded into disaccharides by the enzymes, nutrients are absorbed by the nutrient transporters and passed through intestinal epithelial cells (Ashwell, 2009). The nutrient transporters are expressed at a low level before hatching, but are upregulated after hatching (Yadgary et al., 2011; Wong et al., 2017). During the late-term of incubation, very small amounts of carbohydrates are presented in the intestine, the increase of SI expression at the apical membrane allows more carbohydrates to degrade into glucose (De Oliveira et al., 2009; Speier et al., 2012; Dong et al., 2012a). Maintaining high expression levels of SI in the small intestine can provide

Table 5. Small intestine disaccharidase activities of goslings between embryonic d 23 to d 7 post-hatching[1].

Items	E23	E27	DOH	D1	D4	D7	SEM	*P*-value
Sucrase, $\mu mol \cdot min^{-1} \cdot g^{-1}$ tissue								
Duodenum	0.06^{e}	0.07^{e}	0.68^{d}	0.98^{c}	1.50^{b}	4.06^{a}	1.400	<0.01
Jejunum	0.15^{e}	0.39^{e}	1.12^{d}	2.50^{c}	4.55^{b}	5.53^{a}	2.118	<0.01
Ileum	0.21^{e}	0.30^{e}	0.69^{d}	1.39^{c}	1.71^{b}	4.03^{a}	1.323	<0.01
Maltase, $\mu mol \cdot min^{-1} \cdot g^{-1}$ tissue								
Duodenum	0.24^{e}	0.66^{d}	3.11^{c}	3.77^{b}	4.51^{a}	3.80^{b}	1.688	<0.01
Jejunum	0.78^{e}	1.78^{d}	5.93^{c}	7.10^{b}	8.58^{a}	7.60^{ab}	3.100	<0.01
Ileum	0.73^{e}	1.84^{d}	4.07^{c}	6.68^{a}	5.75^{b}	5.77^{b}	2.310	<0.01

[a-e]Different superscripts within a raw indicate a significant difference ($P < 0.05$).
[1]Age refers to embryonic d 23 (E23), d 27 (E27), day of hatch (DOH; after hatch but before feeding), and d 1 (D1), d 4 (D4), and d 7 (D7) post hatching.

Table 6. Small intestine sugar transporter gene expression of goslings between embryonic d 23 to d 7 post-hatching[1].

Items	E23	E27	DOH	D1	D4	D7	SEM	*P*-value
SGLT-1								
Duodenum	1.00^{e}	2.27^{d}	4.39^{c}	15.46^{a}	8.37^{b}	8.19^{b}	4.920	<0.01
Jejunum	2.22^{c}	2.63^{c}	2.88^{c}	7.25^{b}	8.21^{b}	16.20^{a}	5.043	<0.01
Ileum	0.69^{d}	0.67^{d}	9.05^{a}	7.79^{b}	6.72^{c}	6.49^{c}	3.433	<0.01
GLUT-2								
Duodenum	1.00^{e}	2.06^{d}	2.16^{d}	6.48^{a}	5.50^{b}	2.73^{c}	2.033	<0.01
Jejunum	1.19^{d}	1.79^{d}	1.66^{d}	4.63^{c}	11.73^{a}	5.63^{b}	3.777	<0.01
Ileum	0.29^{d}	0.57^{c}	1.11^{b}	1.16^{b}	1.48^{a}	1.46^{a}	0.469	<0.01
SI								
Duodenum	1.00^{d}	1.36^{d}	2.53^{c}	8.44^{a}	2.57^{c}	3.82^{b}	2.538	<0.01
Jejunum	1.16^{c}	1.31^{c}	1.38^{c}	3.42^{b}	4.64^{a}	3.72^{b}	1.446	<0.01
Ileum	0.87^{c}	0.85^{c}	3.66^{a}	2.32^{b}	2.24^{b}	2.39^{b}	1.000	<0.01

Abbreviations: GLUT-2, glucose transporter-2; SGLT-1, sodium/glucose cotransporter protein-1; SI, sucrase-isomaltase.
[a-e]Different superscripts within a column indicate a significant difference ($P < 0.05$).
[1]Age refers to embryonic d 23 (E23), d 27 (E27), day of hatch (DOH; after hatch but before feeding), and d 1 (D1), d 4 (D4), and d 7 (D7) post hatching.

sufficient substrate supply for the nutrient transporters such as SGLT-1 and GLUT-2 (Dong et al., 2012a). Glucose is the key fuel and the important metabolic substrate, which is absorbed by intestinal epithelium via apically located SGLT-1 and transported to the blood via basolateral membrane expressed GLUT-2 (Mace et al., 2009; Wong et al., 2017). In the present study, we observed that the expression of ileal SGLT-1 and SI genes increased prior to hatching and then decreased while hatching, whereas ileal GLUT-2 expression increased with age (Table 6). Additionally, the expression of SGLT-1, GLUT-2, and SI genes in duodenum gradually increased till d 1 post-hatching and then continued to decrease (Table 6). However, the expression of jejunal SGLT-1, GLUT-2, and SI genes were kept relatively stable during incubation period, after hatching, jejunal SGLT-1 expression increased with age, whereas jejunal GLUT-2 and SI expression continued to increase till d 4 post-hatching and then decreased (Table 6). In chicks, Gilbert et al. (2007) and Sklan et al. (2003) reported that the expression of SGLT-1, GLUT-2, and SI genes increased with age. Similar results were also observed in pigeons and turkeys (Dong et al., 2012a; Weintraut et al., 2016). Additionally, Weintraut et al. (2016) studied the GLUT-2 expression in turkeys during post-hatching period, they found that the expression of GLUT-2 decreased with age. Barfull et al. (2002) observed the SGLT-1 expression in chicks decreased with age during the first week post-hatching. The upregulation of these sugar transporter expressions prior to hatching obviously makes preparations for further digestion of exogenous feed. However, the reason for the downregulation of sugar transporter expression after hatching is still unclear and it seems to be determined by a series of factors. The expression of sugar transporters also differed temporally and spatially in the small intestine. Kaminski and Wong (2018) noted that the expression of sugar transporters in jejunum of chicks was higher than that in duodenum and ileum. Dong et al. (2012a) observed that expression of SGLT-1 and GLUT-2 were the highest in jejunum and ileum of pigeons. In the present study, the highest expression of SGLT-1 was observed in duodenum and jejunum of goslings, followed by ileum. The GLUT-2 expression was highest in jejunum, followed by duodenum and then ileum. The SI expression was highest in duodenum, followed by jejunum and then ileum. Therefore, we considered that jejunum and duodenum were main sites for sugar transportation in goslings.

CONCLUSIONS

We observed that the small intestine developed rapidly throughout pre- and post-hatching, of which jejunum and ileum have a high priority in intestinal development. The villi were preferentially enriched in duodenum and jejunum and its surface area increased with age. The disaccharidase activities increased with age, of which the highest disaccharidase activities were in jejunum. Additionally, sugar transporter genes were upregulated prior to hatching whereas downregulated post-hatching. Higher sugar transportation gene expression was observed in jejunum and duodenum. A rapid development in intestinal morphology, disaccharidase activities, and sugar transporter gene expression around the hatching indicated that goslings have a high carbohydrate digestion and assimilation potential during its early-life, which provided a preparation for further digestion of exogenous feed.

DISCLOSURES

We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome.

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