Postprint: Early Digestive Characteristics of Squabs

Authors: Zhang Xiaoyun, Dong Xinyang, Bu Xingchen, Qi Mingxing, Miao Liping, Zhou Minyao, Zou Xiaoting

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Abstract

This experiment investigated the early developmental characteristics of the digestive tract in squabs by examining the changing patterns of body weight, digestive organ weights, serum protein metabolism, and digestive enzyme activities in pancreatic and small intestinal mucosal tissues at 2–10 days of age. Thirty White Carneau squabs were selected at 2, 4, 6, 8, and 10 days post-hatching (6 squabs per age group) and slaughtered for sampling. The results showed: 1) Age significantly affected ($P<0.05$) early body weight (including yolk sac weight), organ weights and organ indices of proventriculus, gizzard, liver, pancreas, and small intestine; serum total protein, albumin, uric acid, and urea nitrogen contents; total activities of pancreatic amylase, lipase, and trypsin, lipase specific activity, and total and specific activities of small intestinal mucosal disaccharidases (sucrase and maltase) and alkaline phosphatase. Among these, parameters exhibiting linear increases with age included: organ indices of liver, pancreas, and small intestine ($P<0.01$); body weight and organ weights of gizzard, proventriculus, pancreas, liver, and small intestine ($P<0.01$); serum total protein and albumin contents ($P<0.01$); total activities of pancreatic amylase, lipase, and trypsin, and pancreatic lipase specific activity ($P<0.05$); total and specific activities of small intestinal mucosal disaccharidases (maltase and sucrase) and alkaline phosphatase ($P<0.01$). Serum uric acid and urea nitrogen contents and trypsin specific activity exhibited linear decreases with age ($P<0.05$). Organ indices of proventriculus and gizzard showed a pattern of initial increase followed by decrease ($P<0.05$). 2) Intestinal segment significantly affected total activities of maltase and sucrase in small intestinal mucosa ($P<0.05$). 3) The interaction between age and intestinal segment significantly influenced total maltase activity, total sucrase activity, and sucrase specific activity ($P<0.05$). These results suggest that with increasing age, squabs exhibit continuously enhanced protein metabolic capacity and deposition level, rapid development of the digestive system with growth rate exceeding overall body growth, and progressively increasing hydrolytic enzyme activities in the pancreas and small intestinal mucosa, with significant improvement in overall developmental status by 10 days of age. Therefore, the 2–10 day post-hatching period represents a critical stage for the maturation of digestive physiological functions in squabs.

Full Text

Study on Early Digestive Characteristics of Pigeon Squabs

ZHANG Xiaoyun, DONG Xinyang, BU Xingchen, QI Mingxing, MIAO Liping, ZHOU Minyao, ZOU Xiaoting*

(Key Laboratory of Animal Nutrition and Feed Science of Ministry of Agriculture, Feed Science Institute, Zhejiang University, Hangzhou 310058, China)

Abstract

This experiment investigated the developmental patterns of the digestive tract in posthatch pigeon squabs by examining changes in body weight, digestive organ weights, serum protein metabolism indices, and activities of pancreatic and intestinal mucosal digestive enzymes from 2 to 10 days of age. Thirty White Carneau squabs were selected and euthanized at 2, 4, 6, 8, and 10 days posthatch (six squabs per age group) for sample collection. The results showed that: 1) Day of age significantly affected ($P<0.05$) body weight (including yolk weight), organ weights and indices of proventriculus, gizzard, liver, pancreas, and small intestine, serum total protein, albumin, uric acid, and urea nitrogen contents, total activities of pancreatic amylase, lipase, and trypsin, specific activity of lipase, and total and specific activities of intestinal disaccharidases (sucrase and maltase) and alkaline phosphatase. Specifically, indicators showing linear increases with age included: organ indices of liver, pancreas, and small intestine ($P<0.01$); body weight and organ weights of proventriculus, gizzard, pancreas, liver, and small intestine ($P<0.01$); serum total protein and albumin contents ($P<0.01$); total activities of pancreatic amylase, lipase, and trypsin, and specific activity of pancreatic lipase ($P<0.05$); and total and specific activities of intestinal disaccharidases (maltase and sucrase) and alkaline phosphatase ($P<0.01$). Serum uric acid, urea nitrogen contents, and specific activity of trypsin showed linear decreases with age ($P<0.05$). Organ indices of proventriculus and gizzard increased initially then decreased ($P<0.05$). 2) Intestinal segment significantly affected total activities of maltase and sucrase in small intestinal mucosa ($P<0.05$). 3) The interaction between day of age and intestinal segment significantly affected total activities of maltase and sucrase and specific activity of sucrase ($P<0.05$). These results indicate that with increasing age, protein metabolic capacity and deposition level continuously improved, the digestive system developed rapidly with growth rates exceeding body growth, and endogenous pancreatic and intestinal mucosal hydrolase activities progressively increased, with significant improvement in overall development by 10 days

of age. Therefore, the period from 2 to 10 days posthatch represents a critical stage for the maturation of digestive physiological functions in squabs.

Keywords: squab; protein metabolism; digestive enzyme activity; gastrointestinal tract development

Introduction

With continuous improvement in living standards, consumer demand for safe and high-quality livestock products has increased annually. Pigeon meat is delicious, high in protein and low in fat, rich in mineral elements, and has a balanced amino acid composition that is easily digested and absorbed by humans, leading to the popular saying "one pigeon is better than nine chickens" [1]. Large-scale pigeon farming in China began in the early 1980s and has since developed at an annual growth rate of 10%-15%, with continuously expanding industrial scale. Currently, pigeons have become one of the four major poultry species (chicken, duck, goose, and pigeon) [2]. In recent years, although pigeon production has remained stable with slight increases, it still cannot meet domestic and international market demands. Ji Fengyan [3] reported that Guangdong, Shanghai, Hainan, Jiangsu, Hong Kong, and Macau alone require over 100 million squabs annually, primarily White King pigeons, with annual exports of frozen squabs reaching 2,000-5,000 tons. Pigeon farming has become a high-quality modern agricultural project emphasizing environmental protection and efficiency in specialty poultry production.

Squabs are altricial birds that require natural feeding of crop milk secreted by both parents during the early posthatch period, followed by gradual substitution with moistened grain diets for survival [4]. Compared with other poultry, squabs exhibit rapid growth, high weight gain intensity, strong stress resistance, and short feeding cycles [5-6]. Sales et al. [7] reported that the relative growth rate of squabs is 3.79 times that of fast-growing broilers and 1.96 times that of quail. This rapid growth may be attributed to both the pigeon milk secreted by parents and the developmental characteristics of the squab' s digestive tract. Research indicates that pigeon milk contains approximately 11%-13% crude protein and 5%-7% fat [16]. Consequently, squabs develop a unique pattern of digestive physiological development during the early posthatch period. This study aimed to investigate the early developmental characteristics of the digestive tract in squabs by measuring body weight, digestive organ weights, serum protein metabolism indices, and activities of pancreatic and intestinal mucosal digestive enzymes in White Carneau squabs from 2 to 10 days of age, thereby providing theoretical basis for nutritional requirements research and artificial rearing practices to promote healthy development of the pigeon industry.

Materials and Methods

1.1 Experimental Animals and Sample Collection

Experimental squabs were purchased from Wenzhou Aofeng Pigeon Industry Co., Ltd. Newly hatched squabs were naturally fed by parent pigeons housed in three-tier full-step cages (200 cm × 170 cm × 55 cm). Parent pigeons were fed a basal diet (whole grains) formulated according to the American King Pigeon Breeding Management Technical Standard (DB34/T 541-2005). Diet composition and nutrient levels are shown in Table 1 . Feed and water were provided ad libitum throughout the experimental period. The open-type pigeon house utilized natural ventilation and lighting, with temperature maintained at 20-24 °C and relative humidity at 65%-75%.

Six White Carneau squabs were selected at each of 2, 4, 6, 8, and 10 days of age, weighed, blood-sampled, and then euthanized for tissue collection. The proventriculus (contents removed), gizzard (contents removed), liver, pancreas, and small intestine (contents removed, anteriorly cut at the duodenum-gizzard junction and posteriorly at the cecal heads) were collected and weighed. The small intestine was gently opened and rinsed with physiological saline, and mucosa from the duodenum, jejunum, and ileum were scraped and collected in cryovials. Pancreas and small intestinal mucosa samples were stored at -80 °C for subsequent analysis.

1.2 Analytical Methods

1.2.1 Organ Weight and Organ Index Measurement Body weight and weights of proventriculus, gizzard, liver, pancreas, and small intestine were measured, and organ indices were calculated as follows: Organ index (%) = (organ weight / body weight) × 100.

1.2.2 Serum Protein Metabolism Indices Measurement Blood samples were centrifuged at 3,000 r/min for 15 min at room temperature to prepare serum. Serum total protein (TP), albumin (ALB), urea nitrogen (UN), and uric acid (UA) contents were measured using assay kits purchased from Nanjing Jiancheng Bioengineering Institute.

1.2.3 Digestive Enzyme Activity Measurement Duodenal, jejunal, and ileal mucosal samples and pancreatic samples were mixed with physiological saline at a mass/volume ratio of 1:9. Tissue homogenates were prepared in an ice bath and centrifuged at 4 °C, 10,000 r/min for 20 min, and the supernatant was collected for enzyme activity detection. Tissue total protein (TP) content and activities of pancreatic amylase (AMS), lipase (LPS), and trypsin, as well as intestinal mucosal sucrase, maltase, and alkaline phosphatase (AKP) activities, were measured using assay kits purchased from Nanjing Jiancheng Bioengineering Institute.

1.3 Statistical Analysis

Experimental data are expressed as means and SEM. Data were initially organized using Excel. One-way ANOVA in SPSS 20.0 software was used to analyze changes in digestive organ weights, serum protein metabolism indices, and pancreatic digestive enzyme activities. A general linear model (univariate procedure) was used to analyze small intestinal mucosal digestive enzyme activities. Tukey' s test was used for multiple comparisons. $P<0.05$ was considered statistically significant, and $P<0.01$ was considered highly significant.

Results

2.1 Developmental Changes in Body Weight and Organ Weights of Squabs During Early Posthatch Period

Changes in body weight and organ weights of squabs from 2 to 10 days of age are shown in Table 2 . Day of age significantly affected ($P<0.05$) body weight (including yolk weight) and organ weights and indices of proventriculus, gizzard, liver, pancreas, and small intestine. Body weight and all organ weights increased linearly with age ($P<0.01$). Compared with 2-day-old squabs, the body weight and organ weights of proventriculus, gizzard, liver, pancreas, and small intestine in 10-day-old squabs increased by 6.71-fold, 5.50-fold, 8.59-fold, 10.67-fold, 17.55-fold, and 15.3-fold, respectively. The trends in organ indices varied among organs. The organ indices of proventriculus and gizzard increased initially then decreased with age ($P<0.05$), peaking at 4 or 6 days of age. In contrast, organ indices of liver, pancreas, and small intestine increased linearly with age ($P<0.05$). The growth rate of pancreatic organ index was relatively slow. The small intestine organ index increased steadily from 4 to 6 days of age, reaching 2.11 times that of 2-day-old squabs by 10 days of age.

2.2 Developmental Changes in Serum Protein Metabolism Indices of Squabs During Early Posthatch Period

Changes in serum protein metabolism indices of squabs from 2 to 10 days of age are shown in Table 3 . Day of age significantly affected ($P<0.05$) serum total protein, albumin, uric acid, and urea nitrogen contents. Serum total protein and albumin contents increased linearly with age ($P<0.01$). Compared with 2-day-old squabs, serum total protein and albumin contents in 10-day-old squabs increased by 0.95-fold and 1.25-fold, respectively. Serum uric acid and urea nitrogen contents decreased linearly with age ($P<0.01$). The serum uric acid and urea nitrogen contents in 10-day-old squabs were 1.41-fold and 2.67-fold lower than those in 2-day-old squabs.

2.3 Developmental Changes in Pancreatic Digestive Enzyme Activities of Squabs During Early Posthatch Period

Changes in pancreatic digestive enzyme activities of squabs from 2 to 10 days of age are shown in Table 4 . Day of age significantly affected ($P<0.05$) total activities of pancreatic amylase, lipase, and trypsin. Total activities of pancreatic amylase, lipase, and trypsin increased linearly with age ($P<0.01$). Compared with 2-day-old squabs, total activities of pancreatic amylase, lipase, and trypsin in 10-day-old squabs increased by 1.29-fold, 8.64-fold, and 1.89-fold, respectively. Additionally, day of age significantly affected ($P<0.05$) specific activities of pancreatic lipase and trypsin. Specific activity of pancreatic lipase increased linearly with age ($P<0.05$), with 10-day-old squabs showing 5.39-fold higher activity than 2-day-old squabs. In contrast, specific activity of pancreatic trypsin decreased linearly with age ($P<0.05$), with 10-day-old squabs showing 0.23-fold lower activity than 2-day-old squabs.

2.4 Developmental Changes in Small Intestinal Mucosal Digestive Enzyme Activities of Squabs During Early Posthatch Period

Changes in small intestinal mucosal digestive enzyme activities of squabs from 2 to 10 days of age are shown in Table 5 . Day of age significantly affected ($P<0.01$) both total and specific activities of small intestinal mucosal digestive enzymes. Total activities of small intestinal mucosal digestive enzymes increased slowly from 2 to 8 days posthatch, then increased significantly by 10 days of age. Compared with 2-day-old squabs, total activities of sucrase, maltase, and alkaline phosphatase in 10-day-old squabs increased by 10.83-fold, 1.98-fold, and 1.19-fold, respectively, while specific activities increased by 13.64-fold, 2.39-fold, and 1.82-fold, respectively. Intestinal segment significantly affected ($P<0.05$) total and specific activities of sucrase but had no significant effect ($P>0.05$) on total and specific activities of alkaline phosphatase. Total activities of all enzymes were highest in the ileum, followed by the jejunum, and lowest in the duodenum. The interaction between day of age and intestinal segment significantly affected ($P<0.05$) total activities of maltase and sucrase and specific activity of sucrase.

Discussion

3.1 Developmental Changes in Body Weight and Organ Weights of Squabs During Early Posthatch Period

Growth performance and developmental status of squabs directly affect production efficiency of pigeon farming. During the early posthatch period, growth performance of young poultry is visually reflected by changes in organ weights. Hu et al. [5] reported that squabs exhibit rapid early growth, with the fastest weight gain occurring in the second week posthatch. The present study found that squabs developed rapidly during the early posthatch period, with body weight and organ weights showing linear growth trends, consistent with previous reports. The pattern of organ development during the embryonic period not only affects embryo survival and hatchability but also determines posthatch individual development. Chen et al. [8] reported that pigeon embryos (excluding yolk weight) developed rapidly from embryonic day 9 to hatching, with absolute weight increasing from 1.27 g to 11.47 g and relative weight continuously increasing, most rapidly between embryonic days 13 and 15. These results indicate that neural and brain tissues develop rapidly during early embryonic development, while the gastrointestinal system and liver begin rapid development in later stages. Rapid liver development during late embryonic stages serves the need for glycogen synthesis and energy storage. The gastrointestinal system functions in nutrient ingestion, digestion, and absorption [9]. Rapid small intestine development can enhance nutrient absorption and utilization posthatch, thereby promoting squab growth. After hatching, the digestive system faces the challenge of transitioning from endogenous yolk sac nutrition to high-protein, high-fat caseous pigeon milk fed by parents, requiring rapid intestinal development to achieve functional maturity. This study found that within 10 days posthatch, the weight gain rates of liver, pancreas, and small intestine were 1.51-fold, 2.41-fold, and 2.11-fold higher than body weight gain, respectively, with growth rates significantly exceeding body growth, consistent with reports in young poultry and chicks [10].

3.2 Developmental Changes in Serum Protein Metabolism Indices of Squabs During Early Posthatch Period

Protein is an essential nutrient for animal cell and tissue metabolism and a major component of livestock meat, eggs, and milk products. Protein deposition level in squabs depends on the dynamic balance between synthesis and catabolism. Serum total protein and albumin contents are important indicators reflecting protein deposition capacity in livestock, while serum uric acid and urea nitrogen are end products of protein digestion, with uric acid being predominant in poultry [11]. In this experiment, serum total protein and albumin contents changed consistently, increasing rapidly from 2 to 6 days of age then remaining stable. Conversely, serum uric acid and urea nitrogen contents decreased sharply from 2 to 6 days of age then remained relatively stable. These results indicate that with increasing age, squab growth and developmental functions gradually matured, with nitrogen deposition capacity continuously improving.

3.3 Developmental Changes in Pancreatic Digestive Enzyme Activities of Squabs During Early Posthatch Period

The pancreas is an important solid digestive gland in livestock that secretes pancreatic juice containing abundant digestive enzymes participating in intestinal lumen digestion. Pancreatic digestive enzyme activity is a direct indicator of digestive capacity [12]. Dong et al. [13] found that specific activities of pancreatic amylase and trypsin in pigeons increased sharply at 3 and 8 days posthatch, respectively. However, this experiment found no significant change in amylase specific activity and a decrease in trypsin specific activity after 6 days, possibly due to differences in parental diet composition [5]. Sun et al. [14] detected major pancreatic digestive enzyme activities in different intestinal segments, reporting that trypsin activity increased significantly in the duodenum and decreased in the jejunum in later stages, while lipase activity in the duodenum increased then decreased with age, opposite to changes in the jejunum, indirectly reflecting trends in pancreatic digestive enzyme synthesis capacity. This study found that total pancreatic digestive enzyme activities increased sharply at 8 days posthatch, with total activities of amylase, lipase, and trypsin increasing by 1.05-fold, 6.81-fold, and 1.54-fold compared with 2-day-old squabs, respectively, with lipase showing the greatest change.

At hatching, squabs must consume pigeon milk secreted by parental crops to survive, with relatively underdeveloped small intestines. Pigeon milk is rich in lipid nutrients and various growth-promoting factors [15-16]. Additionally, studies have shown that pigeon milk contains abundant digestive enzymes that facilitate early digestion in squabs [17-18]. This experiment demonstrated that endogenous pancreatic digestive enzyme activities were low during early posthatch period but increased linearly with age, with substantial increases at 8 or 10 days posthatch, indicating enhanced digestive enzyme synthesis capacity and significantly improved digestion of nutrients in pigeon milk, gradually adapting to exogenous feed nutrition. These results suggest that insufficient endogenous digestive enzyme synthesis capacity during early posthatch period may be one of the key factors limiting squab growth.

3.4 Developmental Changes in Small Intestinal Mucosal Digestive Enzyme Activities of Squabs During Early Posthatch Period

Macromolecular nutrients hydrolyzed by pancreatic enzymes cannot be directly absorbed by intestinal epithelial cells and require further digestion by mucosal enzymes secreted from intestinal mucosa, such as disaccharidases and alkaline phosphatase, to be broken down into monosaccharides, amino acids, and small peptides [10,19-20]. Zou et al. [21] described developmental patterns of chicken small intestinal mucosal disaccharidases (sucrase and maltase), reporting that activities increased with age in the jejunum and ileum but decreased slightly at 6-7 days posthatch in the duodenum before increasing. Uni et al. [22] reported that specific activities of sucrase and maltase in turkeys increased at 2 days posthatch then decreased to minimum levels at 6-7 days. Unlike other poultry, squabs are altricial birds whose sole food source during the first 7 days posthatch is pigeon milk formed by desquamation of lipid-rich epithelial cells from parental crops. Bharathi et al. [17] studied biochemical composition differences between crop tissue and pigeon milk, finding that pigeon milk contained measurable levels of maltase, sucrase, and alkaline phosphatase, with activities gradually decreasing with age, which substantially enhanced squabs' ability to digest exogenous pigeon milk nutrients. Dong et al. [13] reported that specific activities of squab disaccharidases were lowest in the duodenum, showing an initial increase followed by decrease with age. This experiment demonstrated that day of age, intestinal segment, and their interaction all affected mucosal digestive enzyme activities. Small intestinal mucosal digestive enzyme activities changed linearly with age, increasing significantly at 8-10 days posthatch, with enzyme activity levels higher in the ileum than in the duodenum.

Conclusion

In conclusion: 1) During early posthatch period, the heterochronic growth rate of digestive organs in squabs significantly exceeded body growth, representing an important stage of prioritized digestive system development. From 2 to 6 days posthatch, protein utilization efficiency and deposition capacity improved significantly. 2) Endogenous pancreatic and small intestinal mucosal hydrolase activities were low during early posthatch period but increased linearly with age. Ileal mucosal disaccharidase (sucrase and maltase) activities were significantly higher than those in the duodenum.

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Note: Figure translations are in progress. See original paper for figures.