Effects of Dietary Fat Levels on Nutrient Digestibility at Different Sites of Chicken Intestines

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The present study was conducted to investigate the effects of varying dietary fat levels (3%-10%) on the digestibilities of crude protein, crude fat (CF), nitrogen-free extract and ash at different sites of fistulized chicken intestines. Chickens were fistulized to either the middle part of the jejunum (MJ), the distal end of the jejunum (DJ), the middle part of the ileum, the distal end of the ileum or the distal end of the rectum. Intestinal digesta were collected from each site of intestine, and contents of crude protein, crude fat, nitrogen-free extract and ash were measured. The true digestibility of crude protein in intestinal digesta at MJ in the 10% CF group was significantly lower than that in other groups. The digestibility of crude fat in intestinal digesta at MJ in the 10% CF group was also significantly lower than that in other groups. The digestibility of nitrogen-free extract at MJ and DJ in the 10% CF group was significantly lower than that in other groups. There were no significant differences in digestibility of ash among groups. These results clearly demonstrate that dietary fat levels influence the digestibility of protein, fat, and carbohydrate at MJ in chicken.

Key words: carbohydrate, digestibility, fat, fistula, protein

Introduction

Animal fats and vegetable oils are usually used as feed ingredients because fat is not only an important energy source but also a source of essential fatty acids. Thus, the knowledge of the anatomical site of fat absorption is of basic importance in studying the mechanism of its absorption. Renner (1965) reported that the dietary fat absorption was maximum in the distal part of the small intestine and negligible in the large intestine. The dietary fat was digested to fatty acids between the duodenum and the upper ileum in chickens (Hurwitz et al., 1973; Sklan et al., 1975; Krogdahl, 1985). However, the effects of dietary fat levels on nutrient digestibility at different sites of chicken intestines have not been determined.

There is evidence that the differences of dietary fat source and content affect nutrient digestibility. For example, the different dietary fats, such as tallow, lard and soybean oil, showed different digestibility rates (Renner, 1965). Mateos and Sell (1980) indicated that supplemental fat enhanced the utilization of energy from dietary carbohydrate in laying hens. Thus, it is possible that dietary fat levels influence digestibility of nutrients in chickens. Recently, we investigated the effects of dietary protein levels on the nutrient digestibility using fistulized chickens and found that digestibility of fat elevated with the increase of dietary protein levels in the middle and the distal part of the jejunum (Kamisoyama et al., 2009). It is well known that the primary site for amino acid, glucose, starch and fatty acid absorption is the small intestine in birds (Whittow, 2000). Therefore, this method could be useful to examine the effects of dietary fat levels on nutrient digestibility at different sites of chicken intestines.

In this study, we investigated the effects of dietary fat levels on the digestibilities of crude protein (CP), crude fat (CF), nitrogen-free extract (NFE), and ash using fistulized chickens. Our findings suggest that the dietary fat levels influence the absorption of protein, fat, and carbohydrate in the middle part of the jejunum in chickens.

Materials and Methods

Birds and Surgical Procedure for Fistulation

Fifty 190 day-old male Single Comb Whit Leghorn (Ghen Corporation, Gifu, Japan) were used in this study. The birds were divided into 5 groups of 10 birds each. Each group was fistulized on either the middle part of jejunum (MJ, at the position of 11 cm anterior to Meckel’s diverticulum), the distal end of the jejunum (DJ, at the position of 2 cm anterior to Meckel’s diverticulum), the middle part of the ileum (MI, at the position of 12 cm anterior to ileoceccolicc junction), the distal end of the
ileum (DI, at the position of 3 cm anterior to the ileoecocolonic junction) or the distal end of the rectum (DR, at the position of 1 cm anterior to the cloaca) as described previously (Isshiki et al., 1989). All experimental procedures followed the guidelines for the care and use of experimental animals at the Rokkodai Campus of Kobe University in Japan.

**Postoperative Care**

Fistulized birds were placed into individual cages and fed commercial diet (CP 21%, ME 285 kcal/100 g diet, Nippon Formula Feed Mfg Co., Ltd. Kanagawa, Japan) and water *ad libitum*. Birds that showed a decrease in body weight were tube-fed 10 g of recovery diet (casein 18.00%, glucose 58.02%, corn starch 10.00%, cellulose 2.00%, vitamin mixture 0.20%, mineral mixture 5.63%, choline chloride 0.15% and aluminum silicate 1.00%) for 5 days after operation. Postoperative care and observation were the same as described previously (Isshiki et al., 1989).

**Sample Collection**

Birds had free access to water and the experimental diets with various dietary CF levels (Table 1) for a 3-days adaptation period followed by a 3-days collection of digesta from each site of intestines. Digesta were collected at 9:00 and 17:00, and samples from each site of intestine were pooled, mixed and dried at 55°C for 48 hours.

**Measurement of Feed Passage Time**

One day after the sample collection, the experimental diets which were mixed with 1% carmine were tube-fed to the birds, and the time of the first appearance of the carmine-stained digesta was recorded as the feed passage time.

**Chemical Analysis**

CP, ash, crude fiber and solid in digesta was measured by the standard methods (AOAC, 1995). The CF in digesta was also measured by the standard method of AOAC (1995). Briefly, the CF was extracted by ether from about 10 g of digesta using a soxhlet apparatus and weighed using an electronic digital balance (readability 0.0001 g). The content of CF in digesta was 0.1–0.8 g. The content of NFE was calculated by the subtraction of the contents of CP, CF, ash and crude fiber from the content of solid. Apparent digestibilities of CF and ash were calculated by the method of Rutherfurd and Moughan (2003). The intestinal digesta contains endogenous nitrogen including digestive enzymes, mucopolysaccharides, desquamated cells, urea, amino acids produced by cell catabolism, serum albumin and micro-organisms (Rérat, 1978). Therefore, to calculate the true digestibility of CP, the nitrogen content in digesta of chickens fed the protein free diet (α-corn starch 10.0%, corn oil 3.0%, cellulose 3.0%, mineral mix 5.0%, vitamin mix 1.2%, β-corn starch 76.7%, chromium oxide (III) 0.5%) are subtracted as endogenous nitrogen from the nitrogen content of other groups. For the determination of Cr concentration, feed and digesta were ashed by a wet-ash digestion with sodium molybdate, sulfuric acid and nitric acid. Cr concentration was determined at a wavelength of 440 nm (Bolin et al., 1952).

### Table 1. Composition of experimental diets (g/100 g diet)

<table>
<thead>
<tr>
<th></th>
<th>3%</th>
<th>5%</th>
<th>8%</th>
<th>10%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Casein</td>
<td>15.0</td>
<td>15.0</td>
<td>15.0</td>
<td>15.0</td>
</tr>
<tr>
<td>α-Corns starch</td>
<td>10.0</td>
<td>10.0</td>
<td>10.0</td>
<td>10.0</td>
</tr>
<tr>
<td>Tallow</td>
<td>3.0</td>
<td>6.0</td>
<td>8.0</td>
<td>10.0</td>
</tr>
<tr>
<td>Cellulose</td>
<td>3.0</td>
<td>3.0</td>
<td>3.0</td>
<td>3.0</td>
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<tr>
<td>Mineral mix(^1)</td>
<td>5.6</td>
<td>5.6</td>
<td>5.6</td>
<td>5.6</td>
</tr>
<tr>
<td>Vitamin Mix(^2)</td>
<td>1.2</td>
<td>1.2</td>
<td>1.2</td>
<td>1.2</td>
</tr>
<tr>
<td>β-Corn starch</td>
<td>61.7</td>
<td>58.7</td>
<td>56.7</td>
<td>54.7</td>
</tr>
<tr>
<td>Chromium oxide</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
</tr>
</tbody>
</table>

Calculated content:

- Crude protein (%): 14.67, 14.67, 14.67, 14.67
- Crude fat (%): 3.04, 5.04, 8.04, 10.04
- Nitrogen free extract (%): 74.94, 72.94, 69.94, 67.94
- Crude fiber (%): 2.20, 2.20, 2.20, 2.20
- Crude ash (%): 5.15, 5.15, 5.15, 5.15
- Metabolizable energy (kcal/g diet): 2.85, 2.90, 3.10, 3.30

\(^1\) Provided the following quantities of microminerals per kilogram of complete diet: Cu, 25 mg as copper sulfate; Fe, 120 mg as iron sulfate; I, 0.30 mg as potassium iodate; Mn, 25 mg as manganese sulfate; Se, 0.30 mg as sodium selenite; and Zn, 125 mg as zinc oxide.

\(^2\) Provided the following quantities of vitamins per kilogram of complete diet: vitamin A, 10,032 IU as vitamin A acetate; vitamin D₃, 992 IU as D-activated animal sterol; vitamin E, 88 IU as α-tocopherol acetate; vitamin K₃, 1.52 mg as menadione dimethylpyrimidinol bisulphite; thiamin, 1.5 mg as thiamine mononitrate; riboflavin, 10 mg; pyridoxine, 4.0 mg as pyridoxine hydrochloride; vitamin B₁₂, 0.05 mg; D-pantothenic acid, 25 mg as calcium pantothenate; niacin, 60 mg; folic acid, 1.5 mg; and biotin, 0.4 mg.
**Statistical Analysis**

All data were analyzed using two way analysis of variance (ANOVA). Differences among means of the groups or the sites of intestine were analyzed by the Tukey-Kramer test. All statistical analyses were performed using the commercial package (StatView version 5, SAS Institute, Cary, NC, USA, 1998).

**Results**

As shown in Fig. 1, the nitrogen content in digesta tended to increase at the MI ($p < 0.07$), DI ($p < 0.09$) and the DR ($p < 0.09$) compared to MJ and DJ in the protein free diet group. The true digestibility of CP in digesta was significantly ($p < 0.05$) lower at MJ in the 10% CF group, whereas there was no significant difference at DJ, MI, DI and DR among groups (Fig. 2). Significant interaction was not observed between dietary fat levels and sites of intestine ($p < 0.20$).

Apparent digestibility of CF in digesta tended to elevate along the intestinal tract (3%, $p < 0.07$; 5%, $p < 0.10$; 8%, $p < 0.10$; 10%, $p < 0.15$) in all groups (Fig. 3). Apparent digestibility of CF at MJ in the 10% CF group was significantly ($p < 0.05$) lower than that in other groups. Significant interaction was not observed between dietary fat levels and sites of intestine ($p < 0.18$).

As shown in Fig. 4, apparent digestibility of NFE tended to elevate along the intestinal tract (3%, $p < 0.09$; 5%, $p < 0.11$; 8%, $p < 0.08$; 10%, $p < 0.11$) in all groups. Apparent digestibility of NFE at MJ and DJ in the 10% CF group was significantly ($p < 0.05$) lower than that in other groups. Significant interaction was not observed between dietary fat levels and sites of intestine ($p < 0.22$).

There were no significant differences in apparent digestibility of ash among groups (Fig. 5). Significant interaction was not observed between dietary fat levels and sites of intestine ($p < 0.44$).

As shown in Table 2, feed passage time was influenced by dietary fat levels. Feed passage time at MJ in the 10% CF group was significantly ($p < 0.05$) longer than that in 3% and 8% CF groups. Feed passage time at MI in the 3% CF group was significantly ($p < 0.05$) shorter than that in other groups.

**Discussion**

Nitrogen content in digesta tended to increase at the ileum and the rectum in the protein free diet group (Fig. 1), indicating that products of endogenous nitrogen, such
as digestive enzymes, mucoproteins, desquamated cells, urea, amino acids produced by cell catabolism, serum albumin and micro-organisms (Rérat, 1978), influence the nitrogen content of digesta. The true digestibility of CP in digesta was significantly \((p < 0.05)\) lower at MJ in the 10% CF group (Fig. 2). Hulan and Bird (1972) reported that high fat diet feeding tends to lower the protease activity in pancreatic juice. Thus, the reason for the lowered digestibility of CP at MJ in the 10% CF group might be due to insufficient digestion by pancreatic protease.

In the present study, the apparent digestibility of CF at DI was above 85% in all groups and was similar to that at DR. Our results are good agreement with those of Renner (1965). He reported that the absorption of dietary fat occurred in the small intestine and, to lesser extent, in the rectum. Bile, which is produced and secreted by the liver, plays an important role in fat digestion. For example, dietary supplementation of bile salts increases the apparent fat absorption in chicks (Gomez and Polin, 1976). Lindsay et al. (1969) observed an influence of dietary fat on bile secretion in adult cockerels and found that the inclusion of 15% corn oil or herring oil in a basal diet increases the concentration of bile acids in the excreta by about 20%. In addition, it must be noted that the increase of the dietary fat elevated lipase activity in pancreatic juice (Hulan and Bird, 1972). In the present study, the apparent digestibility of CF at MJ in the 10% CF group was significantly lower than that in the 3, 5 and 8% CF groups. It is not clear why the apparent digestibility of CF at MJ in the 10% CF group was lower than other groups. Since tallow was used as a fat source in this experiment, further study is needed to confirm the effects of dietary tallow levels (3–10%) on the bile secretion and lipase activity in pancreatic juice.

The apparent digestibility of NFE in digesta was above 90% at MI in all groups but the digestibility of NFE in the 10% CF group was significantly \((p < 0.05)\) lower than that in other groups at the jejunum (Fig. 4). Hulan and Bird (1972) reported that high fat diet feeding tended to lower the amylase activity in pancreatic juice. Osman (1982) found that amylase is produced by both the pancreas and the intestine in chickens. Siddons (1969) found that the small intestine of chicks contains disaccharidase. It is therefore possible that digestive enzymes, which were produced by the pancreas and the intestine, were influenced by dietary fat levels.

Mateos et al. (1982) reported that addition of graded levels of fat to practical diets reduces the rate of food passage in laying hens and suggested that supplemental fats might improve digestibility of other dietary constituents, and thereby increased the utilization of dietary energy. In this study, dietary fat levels influenced the feed passage time at MI (Table 2) and did not influence the nutrient digestibilities at the ileum and the rectum (Figs. 2–5). Our findings suggest that supplemental fat enhances the utilization of dietary nutrients by reducing feed passage time at the proximal part of the intestine.

In summary, we investigated the effects of dietary fat levels on the nutrient digestibility of fistulized chickens. Our results showed that digestibilities of CP, CF and NFE were decreased at MJ in chickens fed the high fat diet. These results suggest that dietary fat levels influence the digestion of protein, fat and carbohydrate at MJ in chickens.
Table 2. Effects of fat levels on feed passage time at different sites of intestine (min)

<table>
<thead>
<tr>
<th>Dietary fat level (%)</th>
<th>Middle part of jejunum</th>
<th>Distal part of jejunum</th>
<th>Middle part of ileum</th>
<th>Distal part of ileum</th>
<th>Distal part of rectum</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>23.0 ± 2.5⁰</td>
<td>34.0 ± 3.5</td>
<td>38.5 ± 1.5⁰</td>
<td>279.3 ± 23.2</td>
<td>250.8 ± 34.2</td>
</tr>
<tr>
<td>5</td>
<td>30.7 ± 3.2⁰</td>
<td>55.0 ± 6.2</td>
<td>92.2 ± 9.6⁰</td>
<td>260.8 ± 25.4</td>
<td>298.8 ± 12.1</td>
</tr>
<tr>
<td>8</td>
<td>19.0 ± 5.0⁰</td>
<td>26.5 ± 2.5</td>
<td>93.0 ± 3.0⁰</td>
<td>224.8 ± 47.9</td>
<td>236.5 ± 42.7</td>
</tr>
<tr>
<td>10</td>
<td>46.5 ± 2.5⁰</td>
<td>44.0 ± 5.3</td>
<td>83.0 ± 10.2⁰</td>
<td>285.5 ± 6.6</td>
<td>297.3 ± 62.4</td>
</tr>
</tbody>
</table>

Data represent the mean ± SEM. Means with different superscript are significantly different from each other at each site.

References


Bolin DW, King RP and Klosterman EW. A simplified method for the determination of chromic oxide (Cr₂O₃) when used as an index substance. Science, 116: 634–635. 1952.


