Evaluation of dietary butyric acid supplementation on small intestinal morphology, performance and carcass traits of Japanese quails

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SUMMARY
The present study aimed to investigate the effects of dietary supplementation of different contents of butyric acid on small intestinal morphology, performance, carcass traits of Japanese quails. A total of 384 one day old quails were randomly assigned to 4 dietary treatments with 4 replicates of 24 quails and fed with basal diet including 0, 40, 50 and 60 mg of butyric acid/kg of feed. Growth performance, small intestinal morphology (villus height and width), carcass traits (carcass weight and relative weights of breast, heart, liver and gizzard) were assessed on days 21 and 42. Weight gains and feed conversion ratio were significantly improved in quails supplemented with butyric acid compared to the controls. Carcass and breast yields were also markedly increased in the treated birds. Furthermore, the dietary butyric acid addition has also induced highly significant increases in the jejunal villus dimensions (height and width) since the end of the starting period (day 21) persisting over the growing period (day 42). These results demonstrate that dietary supplementation using high quantities of butyric acid greatly improves the small intestinal morphology by increasing the absorptive surface area that consequently promotes the nutrient assimilation and sustains performance in Japanese quails.

Keywords: butyric acid, dietary supplementation, Japanese quail, growth performance, carcass, jejenum, villus dimensions.

RESUME
Evaluation d'une supplémentation alimentaire en acide butyrique sur la morphologie de l'intestin grêle, la croissance et les caractéristiques de la carcasse chez la caille japonaise

L'objectif de cette étude a été d'étudier les effets d'une supplémentation alimentaire en acide butyrique sur la morphologie de l'intestin grêle, la croissance et les caractéristiques des carcasses chez la caille japonaise. Au total, 384 cailles de 1 jour ont été réparties en 4 groupes égaux (4 sous-groupes de 24 oiseaux) et nourries avec un aliment standard contenant 0, 40, 50 ou 60 mg/kg d'aliment d'acide butyrique. Les paramètres de croissance, la morphologie de l'intestin grêle (hauteur et largeur des villosités) et les caractéristiques des carcasses (poids, et poids relatifs des blancs, du gésier, du foie et du cœur) ont été étudiés aux 21ème et 42ème jours. Les gains de poids et l'efficacité alimentaire ont été significativement accrus chez les oiseaux supplémentés par rapport aux témoins. Les rendements des carcasses et des blancs ont également été augmentés chez les cailles traitées. Enfin, la supplémentation alimentaire en acide butyrique a également induit des augmentations significatives et importantes dans les dimensions (hauteur et largeur) des villosités jéjunales dès la fin de la période de démarrage (21ème jour) qui ont persisté jusqu'à la fin de la période de croissance (42ème jour). Ces résultats démontrent qu'une supplémentation alimentaire par des quantités élevées en acide butyrique permet un développement important de l'intestin grêle conduisant à un net accroissement de la surface d'absorption et de l'assimilation des nutriments ce qui favorise la croissance chez la caille japonaise.

Mots-clés : acide butyrique, supplémentation alimentaire, caille japonaise, croissance, carcasse, jéjunum, dimensions des villosités.

Introduction
Butyric acid is an organic acid with four carbons, generally absorbed from the first part of gut and can have dual effects on the intestine and performance [4]. PRYDE et al. [16] demonstrated that butyric acid plays an important role in intestinal villus growth and it is required for the correct development of the gut associated lymphoid tissue [6]. Butyric acid is also considered as a potential alternative for antibiotic growth promoter in order to ban intensive used of antibiotics and to limit acquired antibioresistance [13]. As confirmed the results of LEESON et al. [13], there is an indication that unlike antibiotics, butyrate helps in the maintenance of intestinal villus structure, compared with the negative effect of antibiotics on them. Also, LEESON et al. [13] stated that butyric acid included in diet up to 0.4% as butyrate glycerides shows no negative effect throughout the 42 days long growth period in broilers. Therefore, butyric acid can be an effective tool to improve intestine growth and consequently to increase the performance of quails. Nevertheless, LEESON et al. [13] also suggested that a mixture of butyrate mono-, di-, and triglycerides seems to provide an alternative route for administering butyric acid via diet of broiler chickens. Because of this, it has been tried to use butyric acid including in mono-, di and triglyceride in the present study. On the other hand, CAVE [5] stated that diet palatability and therefore bird appetite can be influenced by organic acids. All these beneficial actions of butyric acid in particular, make it an organic acid deserving scientific and technical attention as a feed additive to Japanese quails diets.

It was hypothesized that additives such as butyric acid can increase the surface areas of the gastrointestinal tract, leading to improve the absorptive function and secondary the
growth efficiency. Thus, the aim of the present study was to
determine the effects of dietary butyric acid supplementation
on small intestinal morphology, performance and carcass
traits of Japanese quails.

Material and Methods

BIRDS, HUSBANDRY AND PROTOCOL DESIGN

Three hundred eighty-four one day old male Japanese
quails were randomly distributed into 4 equal experimental
groups with 4 replicates of 24 quails. Birds were housed for
6 weeks at a temperature which decreased gradually from 37
to 25°C. Each cage was equipped with automatic drinkers,
and manual self-feeders. Feed and water were provided ad
libitum. There was artificial lighting for 24 hours per day
during the first week, and 23 hours per day from day 8 to day
42. The experimental design was a completely randomized
design.

A standard basal diet meeting the NRC [15] requirements)
and eventually supplemented with butyric acid, was given to
birds (Table I). Control birds were fed with basal diet whereas
in the 3 other groups, butyric acid (including in the positions
1 or 3 in monoglycerides for 25 to 30%, in diglycerides for
50 to 55% and in triglycerides for 15 to 25%, Silo Co, Italy)
was added to the basal diet at 40, 50 and 60 mg/kg of food,
respectively. All animal experimentation was conducted in
accordance with the regulations of Islamic Azad University,
Animal Ethics Committee.

Table I: Composition of the basal diet (ingredient and nutrients) given
to one day old Japanese quails for 6 weeks.

<table>
<thead>
<tr>
<th>Ingredient</th>
<th>g/kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn</td>
<td>497</td>
</tr>
<tr>
<td>Soybean meal</td>
<td>420</td>
</tr>
<tr>
<td>Corn protein powder</td>
<td>35</td>
</tr>
<tr>
<td>Soybean oil</td>
<td>15</td>
</tr>
<tr>
<td>CaHPO₄</td>
<td>7</td>
</tr>
<tr>
<td>Limestone</td>
<td>12.5</td>
</tr>
<tr>
<td>DL Methionine</td>
<td>0.2</td>
</tr>
<tr>
<td>Lysine</td>
<td>0.3</td>
</tr>
<tr>
<td>Premix</td>
<td>10</td>
</tr>
<tr>
<td>Sodium chloride</td>
<td>3</td>
</tr>
</tbody>
</table>

**Calculated composition**

<table>
<thead>
<tr>
<th>Ingredient (g/kg)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein</td>
<td>240.6</td>
</tr>
<tr>
<td>Methionine</td>
<td>4</td>
</tr>
<tr>
<td>Methionine + Cystine</td>
<td>8</td>
</tr>
<tr>
<td>Lysine</td>
<td>13</td>
</tr>
<tr>
<td>Calcium</td>
<td>8.1</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>5.7</td>
</tr>
<tr>
<td>Metabolisable energy (kcal/kg)</td>
<td>847.4</td>
</tr>
</tbody>
</table>

1 Each kilogram contains 550 g of crude proteins and 3 420 kcal
of Metabolisable Energy; 2 supplied per kilogram of diet: vitamin A, 1 650 IU; 
cholecalciferol, 750 IU; vitamin E, 12 IU; vitamin K₃, 1.04 mg; riboflavin, 5 
mg; thiamine mononitrate, 2.04 mg; D-biotin, 15 mg; choline chloride, 4.600 
mg; folic acid, 1.03 mg; niacin, 40.20 mg; calcium pantothenate, 11.09 mg; 
pyridoxine hydrochloride, 3.06 mg; vitamin B₁₂ (cyanocobalamin), 0.3 mg; 
ZnSO₄·H₂O, 71.42 mg; FeSO₄·H₂O, 379.33 mg; CuSO₄·5H₂O, 13.08 mg; Na₂SeO₃, 
0.46 mg; MnSO₄·H₂O, 192.53 mg; KI, 0.41 mg.

**BIRD PERFORMANCE ANALYSIS**

In each cage, bird body weight and food intake were
recorded on days 21 and 42 and thereafter mean body weight
gain, food intake, and food conversion ratio were calculated
for each cage (replicate) between 0 and 21, 22 or 42 days. In
each time period, body weight gain was calculated and
expressed as grams per bird. Food intake (g of food intake/bird)
over the entire grow-out period was calculated by
totalling food consumption in each time interval between
each bird sampling. Food conversion ratio (g of food intake
/g of body weight gain) was calculated by dividing total food
intake by total weight gain in each cage.

**MORPHOMETRIC INDICES OF THE JEJUNUM AND CARCASS
TRAITS**

On days 21 and 42, 8 birds per treatment were randomly
chosen for the determination of carcass traits and
morphometric indices of intestine that includes villus
height and villus width. For that, quails were fasted for
approximately 12 hours and then individually weighed,
killed by cervical dislocation, feathered and eviscerated.
The weights of carcass, breast, liver, heart and gizzard were
recorded and the corresponding percentages (% of live body
weight) were calculated.

Then, the intestine was removed, and tissue samples from
jejenum were taken for morphometric indices evaluation
that included villus height (VH) from the tip of the villus
to the crypt, and villus width (VW; average of VW at one-
third and two-third of the villus) [7]. To measure villus
height and width, 2 cm segments from the middle part of the
jejenum were removed, flushed with physiological saline and
immediately put into a 10% buffered formalin solution until
further processing. After embedding the samples in paraffin,
5 μm sections of each sample were placed on a glass slide and
then stained, using haematoxylin and eosin. A total of 10 villi
per sample (40 villi per treatment) were measured using light
microscope [8].

**STATISTICAL ANALYSIS**

Results were analyzed by ANOVA using the GLM
(General Linear Model) procedure of SAS software (SAS
institute, 2003) [17]. Differences between treatments were
compared by the Duncan’s multiple range tests following
ANOVA, and values were considered statistically different at
p < 0.05. When data were percentages they were transformed
by arc sin square root.

**Results**

Performance data for birds fed butyric acid is detailed
in Table II. Weight gains were markedly increased in quails
supplemented with butyric acid compared to the control
quails during the starting (day 0 to day 21) (p < 0.01) and the

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growing (day 22 to day 42) periods (p < 0.001). Although the differences were not statistically significant, it was also noted that the food intake numerically decreased in treated birds compared to the not supplemented quails. Consequently, food efficiency was significantly improved in butyric acid supplemented quails for the both periods (p < 0.001).

As shown in Table III, the carcass weights and the relative weights of breast were also markedly increased in 42 days old quails fed with butyric acid supplemented diets compared to the controls whereas liver, heart and gizzard relative weights were not significantly altered.

In addition, the villus dimensions (height and width) were dramatically increased in quails supplemented with butyric acid since the end of the starting period (on day 21) (p < 0.01 and p < 0.001, respectively) and this effect in treated quails was also marked when birds were slaughtered at the end of the experimental period (day 42) (p < 0.01) (Table IV).

Discussion

In the present study, it was observed that increases in growth performance and in carcass traits observed in butyric acid supplemented quails were coupled to increases in the jejunal villus dimensions (height and width).

According to LILJA et al. [14], rapid intestinal growth is a prerequisite for sustained body growth rates in birds. In the study of KATANBAF et al. [12], it was reported that the accelerated growth of the gastrointestinal tract after hatching has served the function of the nutrient assimilation.
ZHONGHONG and YUMING [21] observed that the dietary sodium butyrate supplementation at 500 mg/kg increased body weight gain from 0 to 21 days in agreement with the present results. However, LEESON et al. [13] have not found any effect on weight gain during the starting, growing or finishing periods in broilers supplemented with butyric acid. In the same way, HOUSHMAND et al. [9] and HERNANDEZ et al. [10] have also observed no significant effect of organic acid supplementation on growth performance of broilers. ISABEL and SANTOS [11] concluded that the dietary addition of organic acid salts had no effect on body weight and body weight gain in broilers, and that food efficiency was decreased in the supplemented birds compared to the controls. By contrast, slight reduction in the food intake coupled to significant increases in weight gains at the end of the starting and growing periods in butyric acid supplemented Japanese quails observed in the present study have lead to a significant improvement of the food conversion ratio.

On the other hand, villus height and villus width could be considered as indicators for an active functioning of intestine [3]. Increased villus height provides a greater surface area for nutrients absorption and consequently, higher performance [3]. On the contrary, reduction in villus height can reduce nutrient absorption due to the decrease in the intestinal surface area for absorption. Thus, reduction in nutrient absorption, decreased resistance to disease and lower growth performance and increase in secretion of gastrointestinal tract are the negative consequences of deeper crypt and shorter villi [20]. ABDEL-FATTAH et al. [1] indicated that the dietary addition of organic acids increased the weight and length of small intestine. Several studies have previously shown that the addition of butyric acid to broiler diets can have a positive effect on gut morphology and consequently, on performance parameters [2, 6, 18]. For example, LEESON et al. [13] stated that carcass weight and breast meat yield significantly increased in birds fed with 0.2% butyric acid supplemented diet. In agreement with that, Japanese quails supplemented with the organic acid in the present study have also exhibited marked increases in the carcass yield and in breast relative weight compared to the not supplemented controls. On the contrary, VIEIRA et al. [19] indicated that the addition of a blend of organic (lactic, acetic, phosphoric, and butyric) acids did not significantly affect villus height either on days 7, 14, 21 or 42 in broilers.

The discrepancies between studies about the observed effects of dietary supplementation with organic acids, particularly with butyric acid, on growth performances, carcass traits and intestine morphology in poultry may result from the various contents of butyric acid added to the diet and its form (as free acid or included in glycerides) [2]. The unambiguous effects of butyric acid included in glycerides on performance and development of small intestine observed in the Japanese quails in the present study may be related to the high quantities of butyric acid added to the diets that can be extended to other kinds of poultry.

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**References**

DIETARY BUTYRIC ACID ON PERFORMANCES IN QUAILS


