Effect of Butyric Acid Supplementation on Growth Performance and Immune Response in Broilers

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A B S T R A C T

A trial was conducted to investigate the effect of butyric acid on the growth performance and immune response in broiler chicken. 600 day- old broiler chicks were allocated into 4 dietary groups (0, 0.1, 0.2 and 0.4 % butyric acid) with 6 replicates each. Growth performance parameters viz., weekly body weight, feed intake and feed conversion ratio were recorded. Immune response in terms of antibody titer against New castles disease virus, Infectious bursal disease virus and weight of immune organs on day 21 and 42 were measured. The growth performance parameters such as cumulative body weight and feed conversion ratio (FCR) showed significant improvement (P ≤ 0.05) in butyric acid supplemented groups till six weeks and five weeks, respectively. Feed intake was not influenced by the dietary treatments. The immunological responses such as antibody titer, and weights of immune organs (thymus, spleen and bursa of Fabricius) were unaffected on day 21. On day 42 weight of thymus was significantly lower in BA supplemented groups. Butyric acid supplementation could improve the growth performance in broiler chicken.

Keywords
Body weight, Broiler, Feed intake, antibody titer, Growth performance, FCR

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Introduction

Due to growing concerns about antibiotic resistance and the potential ban for antibiotic growth promoters in many countries, there is an increasing interest in finding alternative to antibiotics in poultry production (Patterson and Burkholder, 2003; Chichlowski et al., 2007). The possible alternatives to antibiotics for growth promotion and improvement of feed efficiency in poultry are dietary supplementation of probiotic, prebiotic, feed enzymes, phytobiotics, essential oils and organic acids (Jin et al., 1997; Xu et al., 2006; Biggs et al., 2007). Amongst the organic acids, short chain fatty acids (SCFA) are considered as potential alternative to antibiotic growth promoters (Van Immerseel...
et al., 2005). Butyric acid is a SCFA, which has higher bactericidal activity when the acid is undissociated (Leeson, 2007). Bacterial cell take up undissociated fatty acids and once these acids dissociate, there is change in the intracellular pH leading to death of bacterial cells.

Supplementation of butyric acid derivatives in the diets of broiler chickens could replace antibiotics while maintaining growth performance (Namkung et al., 2011), decrease infection by Salmonella enteritidis (Fernandez-Rubio et al., 2009), and increase growth performance under stress (Zhang et al., 2011).

Materials and Methods

Dietary treatments and management

In the present experiment, 600 day old straight run broiler chicks of uniform body weight were procured from a commercial hatchery. All the chicks were wing banded for identification, weighed and divided randomly into 24 replicates of 25 chicks each. Each of the 4 dietary treatments groups were allotted with 6 replicates. The chicks were reared in open sided house under deep litter system with all standard management practices till 6 weeks of age. Birds were fed (NRC, 1994, Table. 1) with broiler pre starter diet (1-7 days), starter diet (8-21 days) and finisher diet (22-42 days).

Chicks were provided ad libitum supply of feed and water throughout the study. Feeding of test diets commenced from day one and continued till the termination of experiment at six weeks of age. The experimental chicks were vaccinated against Newcastle Disease on day seven with Lasota strain and against Infectious Bursal Disease on day 14 with Intermediate strain through intra ocular route. Booster doses against ND and IBD were given on 21st day and 28th day with F1 and intermediate strains, respectively. All the procedures followed during the trial were approved by the Institutional animal ethical committee, Veterinary College, Bengaluru, KVAFSU.

Growth performance

The body weight of individual birds was recorded at the beginning of the experiment and at the end of each week till six weeks of age to monitor the pattern of body weight gain. The weighing of the birds was done in the early hours of the day before feeding using digital electronic pan topbalance. The daily amount of the concerned diet was weighed and offered separately to each replicate.

At the end of each week, the amount of feed left out in the feeder was removed and weighed carefully without any spillage. Based on the amount of feed given and the left out feed, weekly feed consumption was calculated replicate wise till the end of six weeks. Based on the feed intake per week, the average cumulative feed intake per bird was calculated. The feed conversion ratio (FCR) expressed as the ratio of amount of feed consumed (kg) to the body weight (kg) under each experimental group per week and also on cumulative basis was determined.

Immune response

The serum samples collected on 21st and 42nd days were assayed for antibody titers against Newcastle disease virus (NDV) and Infectious bursal disease virus (IBDV) using Haemagglutination Inhibition test and ELISA, respectively. The micro-test method described by Allan and Gough (1974) was used for detection of HI titer from serum samples collected on 21st and 42nd day to assess the antibody titer.
**Table 1** Per cent ingredient and nutrient composition of basal experimental diet

| Ingredients                        | Prestarter (1-7 days) | Starter (8-21 days) | Finisher (22-42 days) |
|------------------------------------|-----------------------|---------------------|-----------------------|
| Yellow maize                       | 52.17                 | 56.47               | 58.90                 |
| Soya bean meal                     | 40.9                  | 35.54               | 32                    |
| Vegetable oil                      | 3.00                  | 4.3                 | 4.3                   |
| Dicalcium phosphate                | 1.50                  | 1.00                | 0.70                  |
| Common salt                        | 0.40                  | 0.35                | 0.35                  |
| Mineral mixture*                   | 1.50                  | 1.90                | 1.90                  |
| Vitamin premix **                  | 0.20                  | 0.10                | 0.10                  |
| DL-Methionine                      | 0.20                  | 0.15                | 0.15                  |
| Vit.B complex with E***            | 0.05                  | 0.10                | 0.10                  |
| Antibiotic                         | 0.03                  | 0.03                | 0.0                   |
| Coccidiostat                       | 0.05                  | 0.05                | 0.0                   |
| **Nutrient composition**           |                       |                     |                       |
| Crude Protein (%) b                | 22.63                 | 18.92               | 20.45                 |
| Crude Fat (%) b                    | 1.47                  | 4.23                | 4.95                  |
| Crude Fibre (%) b                  | 3.39                  | 3.51                | 3.73                  |
| Moisture (%) b                     | 13.29                 | 10.81               | 10.07                 |
| Ash (%) b                          | 6.86                  | 6.54                | 6.23                  |
| Calcium (%) a                      | 1.03                  | 1.01                | 0.97                  |
| Phosphorous (%) a                  | 0.457                 | 0.453               | 0.424                 |

*Mineral mixture: Each 100 g contains Calcium- 30 %, Phosphorus- 9 %, Manganese- 0.4 %, Zinc-0.4 %, Iron- 2000 ppm, Copper- 500 ppm, Iodine- 100 ppm and Selenium- 23 ppm.

**Vitamin premix: Each gram contains Vitamin A - 82,500 IU, Vitamin B<sub>2</sub> - 50 mg, Vitamin D<sub>3</sub> - 12,000 IU and Vitamin K - 10 mg.

***Vit.B complex with E: Each gram contains Vitamin B<sub>1</sub>- 4 mg, Vitamin B<sub>6</sub>- 8 mg, Vitamin B<sub>12</sub>- 40 mcg, Vitamin E - 40 mg, Calcium D pantothenate- 40 mg and Niacin-60 mg.

a calculated values; b analyzed value

The HI test was done manually by β-procedure in ‘U’ bottom micro-plates using diluters, droppers and 4 HA units of ND viral antigen. The antibody titer against IBDV was measured using Poultry Diagnostic and Research Centre (PDRC) indirect ELISA Kit.

Each of the steps was followed as per the manufacturer’s instructions. The weight of immune organs (spleen, thymus and bursa of Fabricius) of birds supplemented with basal diet and graded levels of butyric acid was measured on 21st and 42nd days of the trial.
Statistical analysis

The data was subjected to One-way analysis of variance (ANOVA) using SPSS statistical software (Version 20 for windows, SPSS). Values were expressed as mean ± SE. Means were compared by Duncan’s test to determine significance between treatments. Significance of difference between treatments means was determined at the P ≤ 0.05.

Results and Discussion

Effect of butyric acid on growth performance

The effects of butyric acid supplementation on growth performance of broiler chickens are presented in Table 2. At six weeks of age, the body weight was significantly higher in butyric acid supplemented groups when compared to the control group.

However, there was no significant difference in body weight among the groups supplemented receiving different levels of butyric acid. Butyric acid supplementation had no significant effect on the feed intake and feed conversion ratio of broilers.

The higher body weight recorded at six weeks of age could be due to improvement in protein and energy digestibility, reducing host nutrients and endogenous nitrogen losses, lowering sub-clinical infections, reducing the production of ammonia and other growth depressing microbial metabolites (Dibner and Buttin, 2002).

In the present study, the improvement in FCR could be attributed to significantly higher body weight due to better intestinal health and high digestibility of nutrients in broilers, which are extremely important in order to attain higher body weight and better FCR (Roberts et al., 2015).

Effect of butyric acid on Immune response

Antibody titer

The effect of butyric acid on the antibody titer against Newcastle disease virus (NDV) and Infectious bursal disease virus (IBDV) on both 21 and 42 days is presented in Table 3. In the present study the antibody titer was not influenced by butyric acid supplemented on both phases of the trial. Imran et al. (2017) supplemented graded levels of micro encapsulated butyric acid (0.025, 0.035 and 0.045 %) and noted no effect on antibody titer against NDV and similar findings were reported by Sikander et al. (2017). Jahanian (2011) reported 0.2 per cent butyric acid glycerides supplementation improved the ND titer at 12th day post vaccination.

Immune organs weight

The effect of butyric acid on immune organs weight as per cent body weight on day 21 and 42 is depicted in Table 4. The weight of spleen and bursa of Fabricius were not affected by dietary supplementation of butyric acid on both phases of the study. The weight of thymus was not influenced by butyric acid supplementation on day 21 and showed significantly lower weight in butyric acid supplemented groups compared to the control group on day 42. Except for the thymus weight, other immune organ weights in the current study were not affected by butyric acid supplementation. This correlates well with the antibody titer against NDV and IBDV which were also not affected with BA supplementation. These findings are in agreement with Mahadavi and Torki (2009) who supplemented butyric acid (0.2 and 0.3 %) in broilers diet and found no effect on immune organs weight and also by Imran et al. (2017) who supplemented graded levels of micro encapsulated butyric acid (0.025, 0.035 and 0.045 %) and found no effect on immune organs was noticed.
### Table 2 Effect of butyric acid Supplementation on body weight in broiler chicken

| Treatment Group                  | Weekly Body weight (g/bird/week) |
|----------------------------------|----------------------------------|
|                                  | I      | II     | III    | IV     | V      | VI     |
| Basal diet (Control)            | 149.03 ± 1.47 b | 386.29 ± 4.24 b | 729.72 ± 8.10 c | 1174.25 ± 11.99 b | 1745.25 ± 19.11 b | 2349.53 ± 25.43 b |
| Basal diet + 0.1% butyric acid  | 152.15 ± 1.64 a,b | 398.52 ± 4.60 a | 770.50 ± 8.99 b | 1250.69 ± 14.53 a | 1801.17 ± 18.44 a | 2422.17 ± 27.59 a,b |
| Basal diet + 0.2% butyric acid  | 153.34 ± 1.34 a,b | 400.41 ± 4.16 a | 766.74 ± 7.40 b | 1255.34 ± 13.13 a | 1825.79 ± 20.37 a | 2458.09 ± 29.81 a |
| Basal diet + 0.4% butyric acid  | 156.09 ± 1.55 a | 407.36 ± 4.24 a | 813.43 ± 8.55 a | 1279.00 ± 13.86 a | 1831.66 ± 19.08 a | 2485.38 ± 25.88 a |
| P value                         | 0.011  | 0.006  | 0.000  | 0.000  | 0.006  | 0.003  |

*Values are means ± standard error. Means within each column with different superscript differ significantly (P ≤ 0.05)

### Table 3 Effect of butyric acid Supplementation on feed consumption in broiler chicken

| Treatment Group                  | Weekly Feed Consumption (g/bird/week) |
|----------------------------------|---------------------------------------|
|                                  | I      | II     | III    | IV     | V      | VI     |
| Basal diet (Control)            | 131.37 ± 4.06 | 554.32 ± 3.12 | 1213.86 ± 17.31 | 2176.88 ± 20.66 | 3211.43 ± 30.77 | 4617.85 ± 37.34 |
| Basal diet + 0.1% butyric acid  | 131.90 ± 2.62 | 564.10 ± 4.80 | 1185.71 ± 25.49 | 2178.10 ± 29.58 | 3232.96 ± 24.51 | 4647.42 ± 39.02 |
| Basal diet + 0.2% butyric acid  | 135.80 ± 3.86 | 559.38 ± 2.16 | 1185.36 ± 25.90 | 2160.73 ± 19.97 | 3212.99 ± 20.43 | 4607.09 ± 31.47 |
| Basal diet + 0.4% butyric acid  | 133.72 ± 3.32 | 557.22 ± 3.06 | 1193.33 ± 10.80 | 2180.16 ± 12.21 | 3214.88 ± 22.78 | 4607.99 ± 37.55 |
| P value                         | 0.806  | 0.258  | 0.746  | 0.914  | 0.921  | 0.848  |

*Values are means ± standard error
Table 4 Effect of butyric acid Supplementation on feed conversion ratio in broiler chicken

| Treatment Group                  | I            | II           | III          | IV           | V            | VI           |
|---------------------------------|--------------|--------------|--------------|--------------|--------------|--------------|
| Basal diet (Control)            | 1.28 ± 0.04  | 1.43 ± 0.03a | 1.66 ± 0.03a | 1.85 ± 0.01a | 1.84 ± 0.02a | 1.96 ± 0.02  |
| Basal diet + 0.1% butyric acid  | 1.25 ± 0.02  | 1.41 ± 0.00ab| 1.54 ± 0.03b | 1.74 ± 0.02b | 1.80 ± 0.01ab| 1.93 ± 0.02  |
| Basal diet + 0.2% butyric acid  | 1.27 ± 0.05  | 1.39 ± 0.01ab| 1.54 ± 0.02b | 1.72 ± 0.01b | 1.76 ± 0.01b | 1.89 ± 0.03  |
| Basal diet + 0.4% butyric acid  | 1.21 ± 0.04  | 1.36 ± 0.01b | 1.47 ± 0.03b | 1.70 ± 0.02b | 1.77 ± 0.03ab| 1.87 ± 0.04  |
| P value                         | 0.700        | 0.161        | 0.004        | 0.001        | 0.122        | 0.232        |

*Values are means ± standard error. Means within each column with different superscript differ significantly (P ≤ 0.05)

Table 5 Effect of Butyric acid supplementation on Antibody titers against New Castle Disease Virus and Infectious Bursal Disease Virus in broiler chicken

| Treatment Group                  | 21st Day     | 42nd Day     |
|----------------------------------|--------------|--------------|
|                                  | Log_{10} NDV titer | IBDV titer | Log_{10} NDV titer | IBDV titer |
| Basal diet (Control)             | 40.66 ± 12.62 | 1453.08 ± 109.58 | 4.66 ± 1.13 | 1632.66 ± 117.16 |
| Basal diet + 0.1% butyric acid   | 19.16 ± 6.56  | 1435.00 ± 64.07 | 3.50 ± 0.74 | 1557.83 ± 125.82 |
| Basal diet + 0.2% butyric acid   | 30.00 ± 10.75 | 1227.83 ± 85.01 | 3.16 ± 0.75 | 1619.83 ± 83.79  |
| Basal diet + 0.4% butyric acid   | 23.00 ± 7.47  | 1510.25 ± 166.53 | 4.00 ± 0.95 | 1571.25 ± 123.79 |
| P value                          | 0.352        | 0.325        | 0.676        | 0.958        |

*Values are means ± standard error
Table 6. Effect of Butyric acid supplementation on Immune organs weight (as per cent of body weight) in broiler chicken

| Treatment Group               | 21st Day               | 42nd Day               |
|------------------------------|------------------------|------------------------|
|                              | Spleen                 | Thymus                 | Spleen                 | Thymus                 | Bursa of Fabricius |
| Basal diet (Control)         | 0.03 ± 0.003           | 0.10 ± 0.009           | 0.14 ± 0.012           | 0.42 ± 0.027<sup>a</sup> | 0.07 ± 0.014      |
| Basal diet + 0.1% butyric acid | 0.04 ± 0.002           | 0.11 ± 0.008           | 0.12 ± 0.005           | 0.32 ± 0.020<sup>b</sup> | 0.07 ± 0.011      |
| Basal diet + 0.2% butyric acid | 0.04 ± 0.003           | 0.12 ± 0.013           | 0.12 ± 0.007           | 0.30 ± 0.025<sup>b</sup> | 0.07 ± 0.012      |
| Basal diet + 0.4% butyric acid | 0.04 ± 0.002           | 0.13 ± 0.013           | 0.14 ± 0.008           | 0.36 ± 0.033<sup>ab</sup> | 0.05 ± 0.003      |
| P value                      | 0.603                  | 0.451                  | 0.319                  | 0.103                  | 0.025             | 0.585             |

*Values are means ± standard error. Means within each column with different superscript differ significantly (P ≤ 0.05)*
From the results of the present study, it can be concluded that the growth performance and feed conversion efficiency in broiler chicken could be improved by supplementation of butyric acid. However, butyric acid supplementation did not show any positive effect on immune response in broilers.

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