The effect of betaine supplementation on performance of laying hens in the tropical climate during the starting period

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Abstract. The study investigated the effects of dietary betaine on the growth performance of laying hens in a tropical climate during the starting period. In total, 192 laying hens strain Lohmann aged three weeks were assigned to four dietary treatments, each consisting of six replicates of eight birds. The dietary treatments included a basal diet (T0) and a basal diet supplemented with betaine at 6 mg/kg (T1), 12 mg/kg (T2), 18 mg/kg (T3). The treatments lasted for eight weeks. Temperature and relative humidity were measured with Hobo-U12, while the wet-bulb temperature was measured using Lutron HT-3027SD. The effect of betaine was not consistent where T3 decreased feed intake (FI) in week 2 but increased FI in weeks 4 and 5. Furthermore, T2 showed higher FI in weeks 6 and 7 than T0 (p<0.05). Accordingly, T2 and T3 led to lower weight gain in week 2, but they compensated with higher weight gain in week 4 when ambient temperature above 30°C occurred for 42 hours in a week. Overall, weight gain and body weight in week 8 was not affected by the treatment. It is concluded that the effect of betaine was more apparent when the birds were exposed to high ambient temperature.

1. Introduction

In addition to heat stress, high environmental temperature fluctuations also cause disturbances in the structure and function of the intestine, including reduced regeneration and integrity of the intestinal epithelium [1], which will suppress the growth rate and efficiency of poultry feed. High-temperature fluctuations often occur in tropical areas such as Indonesia. High temperature is very harmful to livestock, especially the poultry industry.

Temperature fluctuations occur in poultry houses so that less or more feed is consumed than needed so that it affects growth performance [2]. During cold weather, high fluctuation in house temperature can lead to poor feed conversion ratios and health problems [3]. It has been reported that nutrient manipulation can be a viable option to minimize the adverse effects of environmental fluctuations on broilers [4], including supplementation of functional feed additives, such as probiotics, prebiotics, and natural active substances.

Many studies have been conducted to examine the role of betaines in both broiler and laying poultry. Betaine supplementation in broiler rations can increase carcass and breast weight [5,6], increase body weight and ration efficiency [5]. In addition, betaine supplementation in the ration can increase egg production in laying hens and laying quail [6]. This study aimed to investigate dietary betaine's effects on the growth performance of laying hens in a tropical climate during the starting period.
2. Materials and methods

2.1. Animal ethics

The present study was carried out at the Universitas Sebelas Maret, Surakarta, Indonesia. The protocol of this experiment was approved by the Animal Ethical Clearance, Universitas Sebelas Maret.

2.2. Experimental design

In total, 192 laying hens strain Lohmann aged three weeks were assigned to four dietary treatments, each consisting of six replicates of eight birds. The dietary treatments included a basal diet (T0) and a basal diet supplemented with betaine at 0.6 mg/kg (T1), 1.2 g/kg (T2), and 1.8 g/kg (T3). The treatments lasted for eight weeks. The basal diet used was a commercial product, namely PAR-DOC. The betaine was obtained from BETAFIN (anhydrous betaine, 93% purity). The diet was given to the birds in the mash form, and betaine was mixed before feeding. To ensure that the betaine was thoroughly incorporated into the diet, firstly, betaine was mixed with 1 kg of feed, and then the premix was combined with the remaining feed using a mixer.

The cage was considered as the experimental unit. The chickens (n = 24) were weighed daily. The feed consumption was recorded daily based on the cage (n = 24). Bodyweight gains (BWG) and feed intake were then calculated using this information for each phase. Temperature and relative humidity were measured with Hobo-U12, while the wet-bulb temperature was measured using Lutron HT-3027SD. Performance data were analyzed using ANOVA and continued using the DMRT Test.

3. Result and discussion

As shown in Table 1, the effect of betaine in feed intake was not consistent where T3 decreased feed intake (FI) in week 2 but increased FI in weeks 4 and 5. Furthermore, T2 showed higher FI in weeks 6 and 7 than T0 (p<0.05). Weather conditions that often make the behavior of chickens also change. Changes in feeding behavior and changes in behavior stabilize the body condition. In addition, the behavior of chickens, such as scavenging for feed, also makes food scatter. This condition causes the feed given to be not all consumed.

On the other hand, T3 increased FI in weeks 4 and 5 under prolonged heat stress exposure. For instance, He et al. [7] demonstrated that betaine improved Arbor Acres broilers' BWG and feed intake under 32 °C heat stress. Chand et al. [8] found that dietary supplementation of 1.5% and 2% betaine increased the feed intake and BWG and reduced the FCR of fast-growing broilers exposed to heat stress. Similar findings have been reported in the studies of Sakomura et al. [9] and Singh et al. [10], who observed a significant increase in feed intake and BWG of heat-stressed Cobb broilers fed with a diet containing betaine.

Table 1. Feed intake data and duration of exposure above 30°C

| Week | Betaine Levels (g) | Exposure (hours) | P value |
|------|-------------------|-----------------|--------|
|      | 0                 | 0.06            | 0.12   | 0.18   |        |
| 1    | 158.96±11.69      | 155.30±9.49     | 154.16±13.47 | 144.40±9.11 | 15 | 0.196 |
| 2    | 235.66±4.93       | 225.78±8.72     | 239.19±13.21 | 208.32±38.06 | 17 | 0.076 |
| 3    | 281.11±14.89      | 274.98±11.60    | 276.33±17.81 | 283.02±16.40 | 30 | 0.774 |
| 4    | 261.08±37.33      | 241.04±14.21    | 293.07±34.50 | 297.83±36.00 | 42 | 0.017 |
| 5    | 247.48±34.17      | 236.29±47.31    | 285.71±54.95 | 303.08±29.25 | 37 | 0.042 |
| 6    | 337.78±28.84      | 338.46±25.73    | 392.54±45.87 | 345.25±40.94 | 29 | 0.048 |
| 7    | 390.11±39.28      | 371.86±47.85    | 428.91±31.62 | 372.64±34.84 | 29 | 0.064 |
| 8    | 365.19±26.30      | 356.63±56.51    | 403.80±30.20 | 403.80±30.20 | 35 | 0.179 |

Superscript **s** showed a significant difference at p <0.05

Table 3 data show a significant change occurred at week 3 to week 4, where this condition occurred in temperature exposure. Exposure to temperatures above 30°C for 30 hours at week 3, then at week 4, temperatures above 30°C occurred for 42. There was a difference between T2 and T3 (p >0.01).
Our data showed that dietary supplementation of betaine could mitigate the adverse effects of heat stress on feed intake and BWG in laying hens during the starter period, indicating that betaine has potential as an anti-heat stress additive for pullets. The beneficial effect of betaine on the growth performance of heat-stressed poultry is associated with its osmoregulatory, methyl group donors, and antioxidative properties. Betaine is a trimethyl derivative of the amino acid glycine. It is classified as a methyl donor that plays an essential role in the transmethylation process in energy and protein metabolism [11,12]. Betaine can protect cells and microbes in the intestine from osmotic variations while in the digestive tract [10,13].

**Table 2. Bodyweight data and duration of exposure above 30°C**

| Week | Betaine Levels (g) | Exposure (hours) | P Value |
|------|--------------------|------------------|---------|
| 0    | 237.96±16.93       | 0.06             | 0.12    | 0.18    | 15     | 0.994 |
| 1    | 331.87±17.21       | 236.61±11.43     | 238.33±8.60 | 238.56±14.26 | 15     | 0.150 |
| 2    | 431.14±20.84       | 436.63±13.70     | 403.57±23.10 | 381.39±43.779 | 30     | 0.008 |
| 3    | 465.39±20.78       | 462.58±17.35     | 470.64±21.57 | 464.40±35.38  | 42     | 0.949 |
| 4    | 508.59±40.85       | 496.78±33.27     | 510.47±12.90 | 525.95±39.04  | 30     | 0.524 |
| 5    | 648.83±34.93       | 634.33±31.43     | 630.36±14.30 | 640.22±48.56  | 29     | 0.808 |
| 6    | 748.88±40.22       | 731.11±58.14     | 707.03±12.38 | 756.80±36.15  | 29     | 0.063 |
| 7    | 823.05±40.98       | 801.66±61.95     | 860.55±36.02 | 848.19±22.64  | 35     | 0.113 |

Superscript a,b showed a significant difference at p <0.05

**Table 3. Weekly body weight gain**

| Week | Betaine Levels (g) | P value |
|------|--------------------|---------|
| 2    | 94.71±22.55        | 93.73±18.59 | 89.63±10.47 | 74.73±26.84 | 0.329 |
| 3    | 98.47±10.35        | 106.28±8.763 | 75.60±25.42 | 64.44±24.68b | 0.003 |
| 4    | 34.25±15.66        | 25.94±9.377 | 67.07±23.77a | 83.00±28.04a | 0.000 |
| 5    | 41.36±28.89        | 34.20±36.24 | 39.82±23.43 | 61.55±25.79  | 0.405 |
| 6    | 142.07±12.36       | 137.54±15.98 | 119.88±19.93 | 114.26±37.75 | 0.155 |
| 7    | 100.05±62.57       | 96.78±27.70 | 126.16±12.56 | 116.58±15.92 | 0.455 |
| 8    | 74.16±56.04        | 70.55±26.05 | 104.80±30.20 | 91.39±23.09  | 0.321 |

Superscript a,b showed a significant difference with p value <0.05

Figure 1 shows the uniformity data. The objective for flock uniformity is to have 80% of the pullets within plus or minus 10% of the average flock body weight. Flocks with high uniformity have been reported to reach peak egg production earlier and have higher peak production than flocks of low uniformity [14]. On the other hand, poor uniformity is associated with variation in the degree of sexual
maturity of hens, where underweight pullets have delayed onset of egg production [15]. From the data in Figure 3, it shows that Betaine has a positive effect on Uniformity reaching above 80% at T2 and T3.

4. Conclusion

The current results indicated that long-term heat stress induced inferior growth performance and uniformity laying hens during the starter period. Dietary supplementation of betaine was effective in improving growth performance in a long-term duration of heat stress. Supplementation of betaine was effective in improving uniformity.

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