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# Performance, carcass and meat quality of broilers raised at a high stocking density and supplemented with encapsulated *Cosmos caudatus* K. leaf extract

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## ABSTRACT

The aim of this study was to determine the effect of supplementation of encapsulated *Cosmos caudatus* K. leaf extract (ECLE) on performance, carcass and meat quality of broilers at high density pens. Three hundred and seventy broiler chicks were divided into 5 treatments and 5 replications. The treatment was conducted on days 15 to 42 in T0 group (10 chicks/m$^2$ + basal feed), T1 (16 chicks/m$^2$ + basal feed), T2 (16 chicks/m$^2$ + basal feed containing ECLE 0.5 g/kg feed), T3 (16 chicks/m$^2$ + ECLE 1.0 g/kg feed), and T4 (16 head/m$^2$ + ECLE 1.5 g/kg feed). ECLE reduced feed consumption, increased body weight gain, improved feed conversion ratio (FCR), increased the European Production Efficiency Factor (EPEF) and European Broiler Index (EBI) in T2 and T3 (P<0.05). Carcass weight and percentage were increased in T3 and T4 (P<0.05). Breast percentage was higher (P<0.05) in T0, T1 and T2; wing and thigh percentages were higher (P<0.05) in T3 and T4; drumstick and giblet percentages were lower (P<0.05) in T3. Drumstick's pH were higher (P<0.05) in T2, T3 and T4. Thigh fat decreased (P<0.05) at T4. The redness (a*) in T3 and T4 were significantly better. Overall, at high density pens, ECLE at 0.5 g/kg feed enhanced broiler performance, carcass percentage, and breast meat quality.

*Keywords: Cosmos caudatus K., Encapsulation, Stocking density, Performance, Meat quality*

## INTRODUCTION

Broiler chicken (*Gallus gallus domesticus*) is a domesticated chicken that has spread around the world and has become a source of protein (Qanbari *et al.,* 2019). Broilers may grow quickly if they are reared in the proper environment and fed the right nutrition (Jana *et al*., 2015). According to Food and Agricultural Organization (FAO) (2017), more than 66 billion broiler chickens are yearly slaughtered to meet the world's food demands. As the world's population expands, the demand for chicken meat will continue to increase, putting pressure on the poultry industry's efficiency until the need is fulfilled (Hatcher and Lum, 2020).

The stocking density is the number of chickens reared in a certain area (Abudabos *et al.,* 2013). Indeed, high stocking density is one of the method to increase broiler production efficiency. Aside from the efficiency reason, high stocking density will cause oxidative stress and inhibit the broiler growth due to the decreased of feed intake that followed by decrease in productivity (Jabbar and Yousaf, 2017; Abudabos *et al.,* 2013). According to Petek *et al.* (2014), high stocking density causes a decrease in body weight, especially in broilers reared for 42 days.

In addition, the quality of the carcass will also be low when compared to chickens that are not stressed due to high stocking density. In general, the amount of stress experienced by broilers is directly proportional to the length of time they are raised in a high stocking density environment (Goo *et al.,* 2019). Physiologically, broilers exposed to high stocking density will send signals to the hypothalamus, followed by activation of the adrenal gland in anterior pituitary that secretes corticosterone into the bloodstream (Nelson *et al.,* 2018). Osti *et al.* (2017) reported that blood corticosterone increases blood presure and may cause heat shock protein (HSP) resulting in the inhibition of digestive system. This may eventually impair the digestive function and thus nutrient utilization and growth performance of broilers.

In general, stress can be minimized by administration of antioxidants into the feed (Li *et al.,* 2019). Antioxidants can reduce free radical production by reducing reactive oxygen species (ROS) formation with the mechanism of keeping the blood Fe and Cu binding protein to prevent the formation of the new free radicals (Surai *et al.,* 2019). Oxidative stress is the condition where ROS level higher than antioxidant level. Synthetic antioxidants have been routinely used, particularly in stress-exposed broiler chicken. However, the prolonged synthetic antioxidant administration may leave residue in meats that have a carcinogenic effect on broiler meat consumers (Sugiharto *et al*., 2019). Herbal antioxidants are highly recommended to reduce stress in broilers because of the safety of using natural compounds for a relatively long period of time (Sugiharto, 2020). According to Surai (2016), antioxidants may be found in plant extracts, which are effective at reducing oxidative stress. Puvaca *et al.* (2019) further documented that the inclusion of herbal substances improved carcass quality, increased breast muscle percentage, crude protein and decreased abdominal fat of broilers. Herbal treatment to broilers also reduced adipogenesis, resulting in less fat deposition in the body of animals (Santoso *et al.,* 2019).

*Cosmos caudatus* K. is one of the herbal plants containing high antioxidants. Recent literatures shows that antioxidant activity of fresh *Cosmos caudatus* leaves was 46.49% (Tsiompah *et al*., 2021). *Cosmos caudatus* leaves contain phenolic compounds including quercetin (51%), caffeic acid (3.6%) and ferulic acid (3.1%) (Chan *et al.,* 2016). Indeed, Sobel *et al*. (2014) reported that phenolic compounds are very sensitive to environmental factors including light intensity, oxygen levels, humidity and temperature. Other study shows that phenolic compounds can be protected from environmental factors by encapsulation, which is a coating technology that serves to protect, maintain or increase the stability of bioactive compounds. Effective encapsulation method is used to stabilize biological compounds, essential oil and maintain the taste and odor (Vincekovic *et al.,* 2017). One of the microencapsulation techniques is through freeze drying. This method sublimes ice into steam under vacuum after rapid freezing (Yang *et al*., 2020). In most cases, freeze drying is used to dry materials whose bioactive compounds are sensitive to high temperature (Papoutsis *et al.,* 2018).

So far, the application of microencapsulated *Cosmos caudatus* leaf extract has never been practiced to reduce high stocking density induced stress in broilers. Therefore, this study aimed to investigate the effect of supplementation of encapsulated *Cosmos caudatus* leaf extract on performance, carcass and meat quality of broiler chickens reared in high stocking density pens.

## MATERIALS AND METHODS

### Production of Encapsulated *Cosmos caudatus* K. Leaf Extract

The *Cosmos caudatus* K. leaves were purchased from the local market in Semarang city, Central Java Province, Indonesia. The production of encapsulated *Cosmos caudatus* K. leaf extract began with the extraction of *Cosmos caudatus* K. leaves. The *Cosmos caudatus* K. leaves were weighed, washed and dried in a room without direct sunlight. The dried *Cosmos caudatus* K. leaves were ground into powder. The powder was then macerated with 70% ethanol (1:6, g:mL). Leaves powder was soaked in 70% ethanol for 72 hours at room temperature (Karimy *et al.,* 2013). To optimize the extraction process, methanol, which is a highly polar solvent, was used for maceration (Nawaz *et al.,* 2020). The filtrate was obtained by filtering the macerated substance with filter paper. The filtrate was then dried in a vacum rotary evaporator at a maximum temperature of 60°C until the paste was obtained.

Maltodextrin was utilized as a coating material for encapsulation. In a 1:3 (g:mL) ratio, maltodextrin was dissolved in distilled water. In a 1:5 of filtrate to maltodextrin ratio, the maltodextrin solution was then combined with the

*Cosmos caudatus* K. leaf extract filtrate. The leaf extract of *Cosmos caudatus* K. was then freeze-dried to get the powder product. The encapsulated and unencapsulated *Cosmos caudatus* K. leaf extract was then tested for antioxidant activity based on the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) method (Seyedreihani *et al.,* 2017). The test showed that the inhibition percentage of the encapsulated *Cosmos caudatus* K. leaf extract was 74.6%, while unencapsulated *Cosmos caudatus* K. leaf extract was 20.52%.

**Animals and Experimental Diets**

The study used 370 day old chicks (DOC) of broiler MB 202 Lohmann strain with an average body weight of 41.22 ± 0.756 g. At the age of 0-14 days, the chicks were given of commercial crumble feed containing 21-23% crude protein, 5-8% fat, 3-5% crude fiber and 4-7% ash (based on feed label). Drinking water was provided *ad libitum* during the period of experiment. Vaccination was administered at 4 days of age, single dose of active Newcastle disease-infectious bronchitis (ND-IB) vaccine was administered by eye drops and 0.15 ml of inactivated Newcastle disease-avian influenza (ND-AI) was administered subcutaneously. At 14 days, a dose of Gumboro vaccine was also administered through drinking water. The treatments consisted of T0 (10 chicks/$m^2$ and fed on basal diet), T1 (16 chicks/$m^2$ and fed on basal diet), T2 (16 chicks/$m^2$ and supplemented with encapsulated *Cosmos caudatus* K. leaf extract of 0.5 g/kg), T3 (16 chicks/$m^2$ and supplemented with encapsulated *Cosmos caudatus* K. of 1.0 g/kg), T4 (16 chicks/$m^2$ and supplemented with encapsulated *Cosmos caudatus* K. of 1.5 g/kg). The dose of supplementation was based on Karimy *et al.* (2013) and the treatments were applied at the age of 15 to 42 days. The encapsulated *Cosmos caudatus* K. leaf extract was added into the feed (at the end of mixing process). The formulated basal feed is presented in Table 1.

**Performance of Broilers**

Feed consumption, body weight and feed conversion ratio (FCR) were recorded weekly from 15 to 42 days (treatment period). The FCR was calculated as feed consumption (g) / body weight gain (g). EPEF (European Production Efficiency Factor) calculation to determine production efficiency during treatment was calculat-

Table 1. Feed Composition of Broiler at Days 15-42

| | Composition (%) |
|---|---|
| Ingredient | |
| Yellow maize | 57.86 |
| Coconut oil | 2.55 |
| Soybean meal | 34.81 |
| DL-metionine | 0.19 |
| Bentonite | 1.00 |
| Limestone | 1.34 |
| MCP | 1.51 |
| Premix | 0.27 |
| Chlorine chloride | 0.07 |
| Natrium chloride | 0.40 |
| Nutrient Composition | |
| Metabolizable Energy | 3411.00 |
| Crude protein | 23.17 |
| Crude fiber | 2.92 |
| Crude fat | 1.66 |
| Ash | 8.57 |
| Water content | 11.88 |

[1]Metabolizable energy was calculated according to Bolton formula: 40.81 {0.87 [crude protein + 2.25 crude fat + nitrogen-free extract] + 2.5}

[2]Premix contained (per kg of diet) of Vitamin A 7750 IU, Vitamin B1 1.25 mg, Vitamin B2 3.13 mg, Vitamin B6 1.88 mg, Vitamin B12 0.01 mg, Vitamin C 25 mg, Vitamin D3 1550 IU, Vitamin E 1.88 mg, folic acid 1.50 mg, Ca-d-pantothenate 7.5 mg, niacin 1.88 mg, biotin 0.13 mg, Co 0.20 mg, Cu 4.35 mg, Fe 54 mg, I 0.45 mg, Mn 130 mg, Zn 86.5 mg, Se 0.25 mg, L-lysine 80 mg, choline chloride 500 mg, DL-methionine 900 mg, $CaCO_3$ 641.5 mg, dicalcium phosphate 1500 mg.

ed by the formula : (viability (%) × live weight (kg)/age (days) × FCR) × 100. EBI (European Broiler Index) calculation to determine production efficiency during treatment was calculated by the formula : viability (%) × average daily gain (ADG) (g)/ FCR × 10.

**Carcass Characteristics**

Broiler carcass were cut into 8 pieces (breast, 2 thighs, 2 wings, 2 drumsticks and back) The organs (heart, liver, gizzard and abdominal fat) were taken and cleaned using a knife, surgical scissors or tweezers followed by weighing the organs using a digital scale with an accuracy of 0.01 grams. Carcass percentage = (carcass weight/live weight) × 100%. Percentage of carcass piece = (carcass part/carcass weight) × 100%. Abdominal fat percentage = (abdominal fat weight /carcass weight) × 100%. Giblet relative weight = (giblet weight / carcass weight) × 100%.

**Meat Quality**

Meat quality was determined by measuring color, pH and the proximate of the meats. Meat color was determined using a CR-400 chromameter. The chromameter is attached to the sample and left until the scores appears on the monitor. Color analysis uses several parameters including L* (lightness), a*(redness) and b*(yellowness) (Hussein *et al*., 2020). Measurement of pH using a meat pH meter. The calibrated cathode was inserted into the sample and left until the pH scores shown (Lukanov *et al.,* 2015). Proximate meat parameters includes water, protein, fat and collagen content determined using a foodscan. The sample was mashed and placed in a sample cup on the foodscan and the results are displayed on the monitor (Biesek *et al.,* 2020).

**Statistical Analysis**

The data obtained were analysed by one-way analysis of variance (ANOVA) with a significance level of 5% to determine the effect of treatment and if there was a significant effect, it was continued with Duncan multiple range test (DMRT) to determine the differences between treatments. Data was presented by mean. Analysis of the data using SPSS version 16.0.

## RESULTS AND DISCUSSION

Data of total feed consumption, body weight gain, FCR, EPEF and EBI are shown in Table 2. In T2 and T3, dietary encapsulated *Cosmos caudatus* leaf extract decreased cumulative feed consumption (P<0,05) while increasing body weight gain (P<0,05) than in T0, T1 and T4 group, irrespective of the stocking density effect. The FCR at T2 and T3 group were lower than at T0, T1 and T4 group (P<0.05). The EPEF and EBI in T2 and T3 group were higher than in T0, T1 and T4 group (P<0.05). It has widely been documented that stocking density influenced the broiler productivity (Heidari and Toghyani, 2018). The decreased density puts the birds in a more comfortable zone since they have more space to go to the feed and water. On the other hand, high stocking density makes the bird uncomfortable since the area is becoming increasingly limited, making it difficult for them to get feed and water (Gholami *et al.,* 2020). Data in this study showed that high stocking density had no effect on cumulative feed consumption, growth and feed conversion of broilers. These results disagree with the findings of Qaid *et al*. (2016) reporting that increasing stocking density reduced feed consumption and body weight, while increasing FCR of broilers. High stocking density only affect on chicks feed consumption at starter (day 15-21) and grower (29-35) period. Considering the periods of starter and grower periods, the best result of feed consumption were observed for the broilers reared at low density (10 chicks/m$^2$). This is due to the lower stocking density providing more comfort environment, greater physical space and easy access to feeders and drinkers (Henrique *et al.,* 2017). This study also showed that feed consumption of T1 was significantly greater than that of T3 and T4 chicks. Chan *et al*. (2016) reported that *Cosmos caudatus* K. contains some biological compounds such as quercetin, caffeic acid and ferulic acid. These compounds are known for astringent property, which can reduce the passage of digesta in the gastrointestinal tract and therefore reduce feed consumption (Aroche *et al*., 2018).

The current finding indicated that encapsulated *Cosmos caudatus* K. leaf extract at doses of 0.5 g/kg and 1.0 g/kg increased body weight gain. Although having off flavour (Varmaghany *et al.*, 2015), quercetin contained in the *Cosmos caudatus* K. leaves may increase digestibility, leading to the optimum nutrients absorbtion by animals (Biyatmoko *et al.,* 2021). The latter condition may therefore improve the growth performance of broilers. Because feed accounts for more than 70% of production expenses, FCR is the most important element in determining profit in the livestock sector (Osti *et al.,* 2017). According to Lipinski *et al.* (2019), adding herbal feed additive to feed improved all performance metrics, including average daily gain, final body weight, and FCR. Regardless of stocking density, encapsulated *Cosmos caudatus* K. leaf extract reduced the FCR of broiler in the current trial. The findings of this study are consistent with those of Hussein *et al.* (2020), who found that adding phytobiotics to broiler feed reduced FCR to 1.73, compared to 2.03 without phytobiotics. The presence of bioactive components in herbal plants appears to improve protein digestion in broiler's small intestine, resulting in better FCR. In addition to the improved digestion and nutrient utilization, the high content of antioxidant properties in the *Cosmos caudatus* K. leaves may also contribute for the better antioxidantive status and physiological conditions, which may thus improve the growth performance of birds (Ismail *et al*., 2021).

Body weight, FCR, and viability are parameters that influence EPEF score, whereas ADG, FCR, and viability are elements determining the EBI score. The EPEF and EBI of encapsulated *Cosmos caudatus* K. leaf extract treated birds were raised in this experiment. The findings of this study are comparable to that of reported by Puvaca *et al.* (2019), who found that EBI at hens

Table 2. Production Performance of Broilers

| Parameter | T0 | T1 | T2 | T3 | T4 | SEM | P Value |
|---|---|---|---|---|---|---|---|
| Feed consumption | | | | | | | |
| 15-21 | $645^{a}$ | $677^{c}$ | $651^{ab}$ | $656^{ab}$ | $662^{b}$ | 2.99 | 0.001 |
| 22-28 | $813^{a}$ | $949^{d}$ | $882^{c}$ | $850^{b}$ | $881^{c}$ | 9.64 | 0.000 |
| 29-35 | $926^{a}$ | $1166^{c}$ | $996^{b}$ | $957^{ab}$ | $1007^{b}$ | 18.27 | 0.000 |
| 36-42 | $1605^{b}$ | $1258^{a}$ | $1235^{a}$ | $1281^{a}$ | $1375^{ab}$ | 43.77 | 0.03 |
| 15-42 | $3991^{b}$ | $4053^{b}$ | $3767^{a}$ | $3745^{a}$ | $3927^{ab}$ | 38.5 | 0.024 |
| Body weight gain | | | | | | | |
| 15-21 | 493 | 485 | 494 | 477 | 478 | 4.36 | 0.659 |
| 22-28 | 496 | 505 | 511 | 492 | 504 | 7.11 | 0.944 |
| 29-35 | 459 | 413 | 458 | 407 | 475 | 12.16 | 0.304 |
| 36-42 | $527.6^{b}$ | $512.4^{b}$ | $707.6^{a}$ | $751.2^{a}$ | $448.6^{b}$ | 27.59 | 0.000 |
| 15-42 | $1975.4^{b}$ | $1915.4^{b}$ | $2170.8^{a}$ | $2128.2^{a}$ | $1905.6^{b}$ | 28.46 | 0.000 |
| FCR | | | | | | | |
| 15-21 | 0.68 | 0.73 | 0.83 | 0.72 | 0.85 | 0.035 | 0.543 |
| 22-28 | 1.66 | 1.88 | 1.73 | 1.73 | 1.75 | 0.027 | 0.125 |
| 29-35 | $2.04^{a}$ | $2.83^{b}$ | $2.19^{a}$ | $2.41^{a}$ | $2.16^{a}$ | 0.08 | 0.004 |
| 36-42 | $3.06^{b}$ | $2.52^{ab}$ | $1.75^{a}$ | $1.71^{a}$ | $3.23^{b}$ | 0.17 | 0.002 |
| 15-42 | $2.02^{b}$ | $2.12^{b}$ | $1.73^{a}$ | $1.76^{a}$ | $2.07^{b}$ | 0.039 | 0.000 |
| EPEF | $223^{b}$ | $239^{b}$ | $327^{a}$ | $309^{a}$ | $237^{b}$ | 10.86 | 0.000 |
| EBI | $219^{b}$ | $235^{b}$ | $322^{a}$ | $304^{a}$ | $232^{b}$ | 10.72 | 0.000 |

[a,b]Means marked with superscript letters in the same row are significantly different ($P<0.05$)
T0: chicks raised at density of 10 birds/$m^2$ and received no additive, T1: chicks raised at density of 16 birds/$m^2$ and received no additive, T2: chicks raised at density of 16 birds/$m^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 0.5 g/kg, T3: chicks raised at density of 16 birds/$m^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.0 g/kg, T4: chicks raised at density of 16 birds/$m^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.5 g/kg, SEM: standard error of the mean, FCR: Feed Convertion Ratio, EPEF: European Production Efficiency Factor, EBI: European Broiler Index.

Table 3. Carcass Traits of Broiler

| Parameter | T0 | T1 | T2 | T3 | T4 | SEM | P Value |
|---|---|---|---|---|---|---|---|
| Carcass weight (kg) | 1.69[b] | 1.63[b] | 1.75[ab] | 1.85[a] | 1.86[a] | 0.025 | 0.009 |
| Carcass (% LBW) | 69.7[bc] | 69.4[bc] | 66.8[c] | 78.7[a] | 72.1[b] | 0.985 | 0.000 |
| Breast (% CW) | 39.15[a] | 38.88[a] | 39.19[a] | 31.91[c] | 35.28[b] | 0.739 | 0.000 |
| Wing (% CW) | 9.98[b] | 10.05[b] | 10.33[b] | 14.7[a] | 12.9[a] | 0.472 | 0.000 |
| Thigh (% CW) | 15.34[a] | 15.29[a] | 14.95[a] | 12.71[b] | 14.04[a] | 0.265 | 0.001 |
| Drumstick (% CW) | 13.91[b] | 14.05[b] | 14.05[b] | 20.4[a] | 17.98[a] | 0.665 | 0.000 |
| Back (% CW) | 21.60 | 21.71 | 21.47 | 20.13 | 20.38 | 0.231 | 0.068 |
| Giblet (% CW) | 5.29[a] | 5.14[a] | 5.23[a] | 4.65[b] | 4.94[ab] | 0.074 | 0.027 |
| Abdominal fat (% CW) | 2.11 | 1.65 | 1.81 | 1.58 | 1.65 | 0.075 | 0.166 |

[a,b]Means marked with superscript letters in the same row are significantly different ($P<0.05$)
T0: chicks raised at density of 10 birds/m$^2$ and received no additive, T1: chicks raised at density of 16 birds/m$^2$ and received no additive, T2: chicks raised at density of 16 birds/m$^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 0.5 g/kg, T3: chicks raised at density of 16 birds/m$^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.0 g/kg, T4: chicks raised at density of 16 birds/m$^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.5 g/kg, LBW: live body weight, CW: carcass weight, EM: standard error of the mean.

Table 4. pH Value and Chemical Composition of Broiler Meat

| Parameter | T0 | T1 | T2 | T3 | T4 | SEM | P Value |
|---|---|---|---|---|---|---|---|
| Breast | | | | | | | |
| pH | 5.99 | 6.03 | 6.12 | 6.08 | 6.07 | 0.017 | 0.191 |
| Protein | 23.25 | 23.39 | 22.80 | 22.92 | 23.20 | 0.085 | 0.158 |
| Fat | 3.83 | 4.12 | 4.10 | 3.92 | 3.70 | 0.070 | 0.270 |
| Water content | 73.39 | 73.19 | 73.46 | 73.35 | 73.31 | 0.112 | 0.964 |
| Collagen | 1.47 | 1.37 | 1.41 | 1.48 | 1.56 | 0.049 | 0.835 |
| Thigh | | | | | | | |
| pH | 6.22bc | 6.15c | 6.32ab | 6.34ab | 6.36a | 0.023 | 0.007 |
| Protein | 21.63 | 21.33 | 21.49 | 21.50 | 21.43 | 0.106 | 0.949 |
| Fat | 7.54b | 7.51b | 6.42ab | 6.32ab | 5.96a | 0.219 | 0.049 |
| Water content | 73.45 | 72.18 | 72.60 | 72.57 | 73.35 | 0.234 | 0.379 |
| Collagen | 1.57 | 1.69 | 1.67 | 1.62 | 1.8 | 0.054 | 0.778 |

[a,b]Means marked with superscript letters in the same row are significantly different ($P<0.05$)
T0: chicks raised at density of 10 birds/m$^2$ and received no additive, T1: chicks raised at density of 16 birds/m$^2$ and received no additive, T2: chicks raised at density of 16 birds/m$^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 0.5 g/kg, T3: chicks raised at density of 16 birds/m$^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.0 g/kg, T4: chicks raised at density of 16 birds/m$^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.5 g/kg, SEM: standard error of the mean.

supplemented with herbal was more efficient in an economic point of view. Likewise, the FCR, ADG, and break-even production of the herbal supplementation group were higher than the control group, according to Singh *et al.* (2018). In conjunction with this, Aljumaah *et al.* (2020) proposed that feeding herbal supplements to broiler chicks has an AGP-like effect. The dietary uses of plant extract as phenolic sources has been examined as potential tools on improving growth performance, FCR and decline mortality in farm animal (Christaki *et al.,* 2020). The mechanism by which phenolic compounds act as growth promoters includes increasing digestive enzyme secretions (endogenous digestive enzymes, saliva, bile, and mucus) and decreasing the number of pathogenic bacteria in the gastrointestinal tract (GIT) or modulating gut morphology due to their antioxidant and anti-inflammatory functions (Valenzuela-Grijalva *et al.,* 2020). In contrast, the group of birds fed 1.5 g/kg *Cosmos caudatus* K. leaf extract had worse FCR and performance index (EPEF and EBI). *Cosmos caudatus* K. leaf extract in higher concentrations may have harmful effects or serve as a prooxidant that may damage the intestinal mucosa leading to decreased nutrient absorbtion (Surai, 2013). However, such inference should be interpreted with caution as we did not measure the indicator of oxidative stress such malondialdehyde (MDA) in the current study.

The data on carcass are shown in Table 3. In this study, carcass weight were higher (P<0.05) at T3 and T4 than at T0, T1 and T2. Carcass percentage at T3 was higher (P<0.05) than T0, T1, T2 and T4. The T0, T1 and T2 breast percentages were higher (P<0.05) than T3 and T4. The percentage of wings T3 and T4 were higher (P<0.05) than T0, T1 and T2. The percentage of thigh at T3 was lower (P<0.05) than at T0, T1, T2 and T4. T3 and T4 drumstick percentages were higher (P<0.05) than T0, T1 and T2. The percentage of giblet in T3 was lower (P<0.05) than in T0, T1 and T2 but not signifficantly different than T4. High stocking density had no effect on carcass weight and percentage, breast, wings, thigh, or drumstick according to this investigation. This study was in line with Uzum and Toplu (2013) showing that carcass weight was not affected by stocking density. In their study, chickens at low stocking density had a higher dressing presentage, but no significant difference was found. The previous and our current study had contradiction with Madilindi *et al.* (2018), who claimed that a higher stocking density (40 $kg/m^2$) resulted in a lower carcass percentage. Similar to our results, many authors also found no significant differences in carcass percentage at various stocking densities (Tong *et al.,* 2012; Vargas-Rodriguez *et al.,* 2013). Previous study by Oleforuh-Okeleh *et al.* (2014) documented that broilers receiving phytobiotics in feed had higher carcass production. In this study, we discovered that supplementing broilers with encapsulated *Cosmos caudatus* K. had beneficial effects on the breast, thigh, and another carcass components. In Ross 308 broiler, Khattak *et al.* (2014) discovered a similar impact. It seems that the bioactive compounds in the herbs enhanced the availability of essential nutrients, assisting in the digestion and absorption of dietary amino acids, and thus promoting muscle deposition (Raphael *et al.* 2017). The results showed that at doses of 1.0-1.5 g/kg diet, encapsulated *Cosmos caudatus* K. leaf extract reduced the percentage of giblets. In contrast, Hassan *et al.* (2016) found that supplementing flavanoid-containing herbal feed additives (*Moringa oleifera*) had no effect on the liver, heart, and abdominal fat.

The meat pH and proximate data are shown in Table 4. The pH of thigh meat of T4 was higher (P<0.05) than T0, T1, T2 and T3. Encapsulated *Cosmos caudatus* K. leaf extract had a significant effect on thigh fat content, in T4 were greater (P<0.05) than other groups. Stressed animals has been generally associated with the lower

Table 5. Meat Color

| Parameter | T0 | T1 | T2 | T3 | T4 | SEM | P Value |
|---|---|---|---|---|---|---|---|
| Breast | | | | | | | |
| L* | 52.23 | 52.82 | 51.41 | 51.97 | 52.87 | 0.626 | 0.165 |
| a* | 1.85[a] | 1.44[a] | 2.46[ab] | 5.73[bc] | 7.56[c] | 0.677 | 0.004 |
| b* | 10.13 | 9.39 | 10.15 | 9.53 | 12.33 | 0.520 | 0.411 |
| Thigh | | | | | | | |
| L* | 51.81 | 51.55 | 52.53 | 52.45 | 52.01 | 0.588 | 0.086 |
| a* | 6.68[b] | 4.21[ab] | 1.78[a] | 1.92[a] | 1.75[a] | 0.561 | 0.006 |
| b* | 11.18 | 9.03 | 9.13 | 9.68 | 11.35 | 0.373 | 0.114 |

[a,b]Means marked with superscript letters in the same row are significantly different (P<0.05)
T0: chicks raised at density of 10 $birds/m^2$ and received no additive, T1: chicks raised at density of 16 $birds/m^2$ and received no additive, T2: chicks raised at density of 16 $birds/m^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 0.5 g/kg, T3: chicks raised at density of 16 $birds/m^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.0 g/kg, T4: chicks raised at density of 16 $birds/m^2$ and received encapsulated *Cosmos caudatus* K. leaf extract at 1.5 g/kg, SEM: standard error of the mean, L*: Lightness, a*: redness, b*: yellowness.

meat pH, while non-stressed animals prior to slaughter have a higher meat pH (Patria *et al.,* 2016). Encapsulated *Cosmos caudatus* K. leaf extract effectively raised thigh pH and lowered thigh fat regardless of stocking density. Broilers meat pH is influenced by the stress level of the chicken both prior to and at the time of slaughter. In general, bioactive components in herbal products were able to reduce stress condition in broiler, and hence increase the pH values of meats (Kothari *et al.,* 2019). The results demonstrated that independent of the stocking density, the encapsulated *Cosmos caudatus* K. leaf extract reduced thigh fat content. The phenolic components of phytobiotics can increase the level of polyunsaturated fatty acids (PUFA) due to peroxidation activity (Torquato *et al.,* 2018). In general, the increased physical activity and fat deposition had a negative correlation. Physical activity significantly increased lypolisis, mainly in active muscles than inactive muscle (Patria *et al*., 2016). In general, oxidation is favoured by the presence of PUFA in meat. Double bonds of PUFA function as ideal initiators for the oxidation leading to the formation of hydrperoxides and free radicals. Reducing meat fat content was caused by the mechanism of the phenolic compound by lipid reducing activity on *de novo* lipid biosynthesis accompanied by the downregulation of genes involved in fatty acid elongation, such as acdh-1 and acdh-2 (Kalogianni *et al*., 2020).

The meat colour scores are shown in Table 5. The T3 and T4 group showed that the score of redness (a*) of the breast meat was higher ($P<0.05$) compared to that of T0, T1 and T2. No significant effect was observed on thigh redness scores at all treatment groups in this study. Previously, Karunanayaka *et al.* (2016) explained that the lightness values (L*) was related to the pH values of meat. Stress level might develop of pale soft exudative (PSE) meat because of acceleration of postmortem metabolism and biochemical processes. The colour of broiler meat is influenced by several factors including genetics, feed, handling, heat stress, cold stress, environmental stress and the slaughter process (Nasir *et al.,* 2017). Stocking density can be the one of factor that affects broiler meat colour. According to the previous study, a reduction of the stocking density led to an increase in a* value (Simitzis *et al.,* 2012). This supports the research conducted by Eratalar *et al.* (2020) showing that the a* values of meat were higher in the ducks reared at low stocking density, meaning that reducing stocking density is associated with more reddish meats. Therefore, it can be inferred that increasing the stocking density resulted in the lighter colour of the meat. Zhang *et al*. (2012) stated that broiler chickens exposed to stress had a decreased a* score because there was more oxidized myoglobin in their muscles. Further, Wideman *et al.* (2016) explained that myoglobin levels in breast muscles were lower than in thigh muscles. Tang *et al*. (2013) added that the low value of meat redness is dependent on consistently low pH values, which contributes to the unusual redox reactions of myoglobin and haemoglobin. In agreement, Kamboh *et al.* (2013) and Hernandez-Coronado *et al.* (2019) reported that supplementation of phenolic compound can increase the meat redness score. This due to the mechanism of polyphenols that can reduce metmyoglobin (MetMb) to oxymyoglobin ($MbO_2$). The antioxidant polyphenols having a catechol substructure can effectively reduce MetMb to $MbO_2$ with chemical assistance from nucleophilic reactive thiol compounds (Miura *et al.,* 2014).

## CONCLUSION

Dietary administration of encapsulated *Cosmos caudatus* leaf extract at 0.5 g/kg improved performance, carcass characteristics and meat quality parameters of broiler chicken reared at high stocking density.

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