The potential of extract of *Zingiber zerumbet* (L.) Smith as a feed additive to improve the production performances and meat nutritional composition of broiler chickens

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**Abstract.** This study aimed at replacing the use of antibiotic growth promoters (AGP) as the feed additive with the *Zingiber zerumbet* (L.) Smith extract (ZZE) to control Salmonellosis in broiler chickens. This study employed the completely randomized design (CRD), five treatments, namely: T0 (basal diet), T1 (basal diet + *S. enteridis* infection), T2 (basal diet + *S. enteridis* infection + 0.33 % ZZE), T3 (basal diet + *S. enteridis* infection + 0.67 % ZZE), and T4 (basal diet + *S. enteridis* infection + 1 % ZZE). The addition of the *Z. zerumbet* extract did not seem to affect the production performances, in which the average feed consumption was 69.38 g ± 4.43 g to 72.71 g ± 4.92 g, body weight gain was 35.67 g ± 1.40 g to 38.30 g ± 3.85 g, feed conversion was 1.89 ± 0.11 to 2.04 ± 0.11, final weight was 1 287.80 g ± 49.97 g up to 1 380.50 g ± 134.90 g, and percentage of carcass was 60.90 % to 68.48 %. However, the use of *Z. zerumbet* extract with the highest concentration at 1 % decreased the crude fat of meat.

**Keywords:** Herbal plant, phytobiotics, antibiotic growth promoters, bacterial resistance.

1. Introduction

The increasing prevalence of Salmonellosis caused by *Salmonella enteritidis* pathogenic bacteria that are transmitted vertically or horizontally [1, 2] in broiler chickens has caused serious problems for the poultry industry due to the loss instigated by growth disturbance, decreasing performance, growing number of rejected chickens, as well as increasing sensitivity of chickens to other diseases [3]. Salmonellosis also means a lot to the public health as the livestock products contaminated with *Salmonella* sp. may cause foodborne disease in human; thus, the products must be destroyed. This eventually results in the loss of many business opportunities in the poultry industry [2]. Meanwhile,
the prevention of Salmonellosis with the use of antibiotic growth promoters (AGP) as feed additives in fact may arouse greater problems, such as increasing resistance to antibiotics, increasing outbreaks of Salmonellosis, bacterial contamination in either poultry products and their surrounding environments, foodborne diseases, as well as antibiotic residues in products which may threaten the public health [4–6].

The potential of herbal plants (phytobiotics) in Indonesia is that they may become an alternative of natural AGP substituents and be used as green products to prevent from Salmonellosis in broiler chickens. One of the phytobiotics that are often used to maintain the livestock health is *Zingiber zerumbet* L. Smith as it contains antibacterial properties [7, 8]. The crude ethanol extracts and their fractions of *Z. zerumbet* has antibacterial activity against negative bacteria, so the extract can be used as a natural feed additive [9].

The specific aim of the study was to find out the right concentration of ethanol to extract the antibacterial substances in *Z. zerumbet* as well as the precise concentration of the extract as feed additives that could replace AGP to prevent the livestock from Salmonellosis caused by *S. enteridis*. Therefore, the improvement of production performance and health of broiler chickens for testing the safety of the *Z. zerumbet* extract as feed additives in broiler chickens can be accomplished. As results, it is essential to conduct a series of experiments in whether or not giving the *Z. zerumbet* extract as feed additives affects the activity of digestive enzymes, production performance and quality of broiler chicken meat which artificially infected with *S. enteridis*.

In the long run, the use of phytobiotics as feed additives is expected to increase the productivity of broiler chickens, so that Salmonella-free broiler meat supply is maintained to meet the domestic demands. Besides, one more important target of using phytobiotics as feed additives is to reduce the level of bacterial resistance to antibiotics, suppress foodborne diseases, and antibiotic residues in both meat and visceral, hence it would contribute to the food security and safety in Indonesia.

2. Materials and methods

2.1. Plant materials and preparation

The *Z. zerumbet* rhizome was obtained from Unit Pelaksana Teknis (UPT) Materia Medica Batu, East Java, Indonesia. This rhizome was initially sort out in order to separate it from any foreign material; then it was washed, drained, sliced, and dried in an oven with a temperature of 45 °C until the water concentration reached ≤ 10 %. Next, the dried rhizome was ground into powder, soaked with 95 % ethanol in a jar and stirred at a speed of 50 rpm for 24 h (1 rpm = 1/60 Hz). The liquid extract was then filtered, vaporized with a rotary evaporator for 1 h and above a water bath for 12 h. At the last step, the thick liquid extract was supplemented with aquadest so that it became a 10 % extract. Finally, the extract was dried in an oven at 40 °C for 2 d and added with 5 % amylopectin starch until it became powder.

2.2. Diet management

The feed formulation of the broiler chickens is shown in table 1.

| Feedstuff  | Starter Amount (%) | Finisher Amount (%) |
|------------|--------------------|---------------------|
| Rice bran  | 0.6                | 0.4                 |
| Yellow corn| 59.22              | 65.11               |
| Fish flour | 2.8                | 2.31                |
| Bone flour | 1                  | 2                   |
| Meat flour | 4.42               | -                   |

Table 1. Feed formulation of the treated broiler chickens.
### Feed formulation

| Feedstuff   | Starter | Finisher |
|-------------|---------|----------|
|             | Amount (%) | Amount (%) | |
| Soybean pulp | 29       | 27       | |
| Cocunut oil  | 1.22     | 1.12     | |
| Calcium      | 0.35     | 0.5      | |
| Salt         | 0.5      | 0.5      | |
| Lysine       | 0.7      | 0.9      | |
| Metionine    | 0.19     | 0.16     | |
| **Total (%)**| **100.00**| **100.00**| |

#### Table 1. Continued.

| Feed Composition | Metabolic Energy (kcal kg⁻¹) | Crude Protein (CP) (%) | Crude Fat (CF) (%) | Crude Fiber (CFB) (%) | Ca (%) | P (%) | Na (%) | AMINO ACIDS |
|------------------|-----------------------------|------------------------|-------------------|----------------------|--------|-------|--------|-------------|
|                  | 3000.00                     | 23                     | 4.00              | 3.55                 | 0.89   | 0.42  | 0.15   | 0.88        |
|                  |                             |                        |                   |                      |        |       |        | 0.35        |
|                  |                             |                        |                   |                      |        |       |        | 0.77        |
|                  |                             |                        |                   |                      |        |       |        | 0.98        |
|                  |                             |                        |                   |                      |        |       |        | 0.99        |
|                  |                             |                        |                   |                      |        |       |        | 0.46        |
|                  |                             |                        |                   |                      |        |       |        | 0.76        |
|                  |                             |                        |                   |                      |        |       |        | 0.66        |
|                  |                             |                        |                   |                      |        |       |        | 0.31        |
|                  |                             |                        |                   |                      |        |       |        | 1.23        |

#### 2.3. Dosage test for Salmonella enteridis infection

It was conducted through a series of in vivo experiments in a form of *S. enteridis* infection with multilevel doses, i.e. $10^{4}$, $10^{6}$, $10^{8}$, $10^{10}$ dan $10^{12}$ in 10-day-old broiler chickens. Then, the clinical symptoms of the chicken were observed for 1 wk after the infection. The expected clinical symptoms were those that showed moderate diarrhea symptoms.

#### 2.4. Care management

The research material consisted of 125 one Day-Old-Chick (DOC) broilers, 125 fish heads of which the initial body weight ranged from 34 g to 47 g, with an average of 39.536 ± 2.481 g and a raising period of 35 d. Then, the ND vaccination was performed on chickens aged 4 d, 17 d and 24 d. Meanwhile, artificial infections using *Salmonella* sp. orally was given on day 10 with optical density (OD) $1 \times 10^{10}$ CFU, while the *Z. zerumbet* extract powder was given when the chickens were 7-day-old to 35-day-old.

#### 2.5. Research design

The study used an experimental method, Completely Randomized Design (CRD), consisting of five treatments, namely: T0 (chickens fed on basal diet), T1 (chickens fed on basal diet + *S. enteridis* infection), T2 (chicken fed on basal diet + *S. enteridis* infection + 0.33 % *Z. zerumbet* extract of the total feed), T3 (chickens fed on basal diet + *S. enteridis* infection + 0.67 % *Z. zerumbet* extract of the total feed), and T4 (chickens fed on basal diet + *S. enteridis* infection + 1.00 % *Z. zerumbet* extract of the total feed).
total feed), and T4 (chickens fed on basal diet + S. enteridis infection + 1 % Z. zerumbet extract of the total feed). Each treatment was repeated five times and each replication consisted of five chickens.

2.6. Measured variables

The variables measured were the production performances (feed consumption, body weight gain, feed conversion, and carcass percentage) and the composition of meat nutrition (dry matter, crude fat, organic matter, crude protein and water). The production displays were obtained by measuring the weight, meanwhile the meat compositions were attained through a proximate analysis.

2.7. Data analysis

The data were tabulated in the Excel Program; the production display data and meat composition were analyzed using analysis of variance (ANOVA). If there were differences among the effect of each treatment, the Duncan Multiple Range Test (DMRT) was then applied.

3. Results and discussion

3.1. The production performance of broiler chickens

The results showed that the use of Z. zerumbet extract powder in feed did not affect the production of broiler chicken, as indicated in Table 2.

| Treatment | Feed Consumption (g chick\(^{-1}\) d\(^{-1}\)) | Feed Conversion | Daily Weight Gain (g chick\(^{-1}\) d\(^{-1}\)) | Final Weight (g) | Carcass Percentage (%) |
|-----------|---------------------------------------------|-----------------|----------------------------------------------|-----------------|------------------------|
| T0        | 72.17 ± 5.34\(^a\)                          | 1.89 ± 0.12\(^a\) | 38.30 ± 3.85\(^a\)                           | 1380.50 ± 134.90\(^a\) | 65.60 ± 1.30\(^a\)   |
| T1        | 71.51 ± 3.62\(^a\)                          | 1.89 ± 0.11\(^a\) | 37.91 ± 1.20\(^a\)                           | 1374.40 ± 47.98\(^a\) | 68.48 ± 8.37\(^a\)   |
| T2        | 71.72 ± 4.20\(^a\)                          | 1.96 ± 0.10\(^a\) | 36.50 ± 0.70\(^a\)                           | 1316.80 ± 24.81\(^a\) | 63.70 ± 4.53\(^a\)   |
| T3        | 69.38 ± 4.43\(^a\)                          | 1.93 ± 0.12\(^a\) | 35.91 ± 2.22\(^a\)                           | 1297.30 ± 77.59\(^a\) | 64.74 ± 0.72\(^a\)   |
| T4        | 72.71 ± 4.92\(^a\)                          | 2.04 ± 0.10\(^a\) | 35.67 ± 1.40\(^a\)                           | 1287.80 ± 49.97\(^a\) | 60.90 ± 3.47\(^a\)   |

Note: Number which are followed by similar alphabet in the same column are similar during Duncan test of 5 %. Alphabet a means there is no differences of feed consumption, feed conversion, daily weight gain, final weight and carcass percentage between treatments.

The addition of Z. zerumbet extract did not affect the production display because the given dose of S. enteridis infection (10\(^{10}\) CFU per mL) was mild, with low virulence, and according to the vaccine dose. Therefore, this condition induces the increase in the body's defense mechanism of the T1 treatment, which caused the production performance similar to those of the control group (T0). This is in accordance with the report that S. enteridis vaccine containing 10\(^{11}\) CFU per mL bacteria is able to improve the body's defense system [3].

The content of curcumin and essential oil in Z. zerumbet extract (T2 to T4) has not been able to overcome S. enteridis (T1) infection, so it is not optimal in stimulating bile and pancreatic secretions to digest carbohydrate, fat, and protein feed ingredients, so it does not affect feed consumption and the production performance of broiler chickens. The results of this study differ from previous studies which proved that curcumin in Z. zerumbet extract was shown to increase bile and pancreatic secretion, thereby increasing feed consumption [10, 11]. In addition, essential oils in Z. zerumbet can increase the digestive performance, relax the small intestine by reducing peristalsis movement of the small intestine, so that the ingesta may stay longer in the small intestine and the process of nutrition
absorption may take place maximally [12, 13]. Flavonoids, terpenoids and saponins are antibacterial agents which can suppress the growth of pathogenic bacteria, consequently the better nutrition absorption and increasing growth and final weight of slaughtered chickens can be achieved [14‒16]. The compounds of Z. zerumbet, including flavonoids, alkaloids, tannins, phenols, polyphenols, terpenoids and essential oils are antibacterial and anti-inflammatory substances [14, 17‒19].

The feed nutrients, which consist of: ME, CP, CF, CFB, Ca and P, in experimental chickens meet the needs of chickens and have the same range between each treatment, so that the achievement of the production display, is almost similar. Additionally, The content of each CF, CFB, protein and ME in feed are according to standards, as stated that the need for CF in broiler chicken feed is at most 8.0 %, maximum CFB is 6.0 %, protein in the starter period is 23 % and 20 % in the finisher one, and energy is 3 200 cal g⁻¹ [20].

3.2. Meat nutrients

The compositions of meat nutrients in the broiler chickens are presented in table 3.

| Treatment | Compositions of meat nutrients |
|-----------|-------------------------------|
|           | Dry Matter (DM) (%) | Crude Fat (CF) (%) | Organic Matter (OM) (%) | Crude Protein (CP) (%) | Water (%) |
| T0        | 25.78 ± 0.61ᵃ | 1.83 ± 0.74ᵃ | 22.93 ± 0.79ᵃ | 22.86 ± 0.87ᵃ | 74.22 ± 0.61ᵃ |
| T1        | 25.69 ± 0.51ᵇ | 1.21 ± 0.39ᵇ | 22.91 ± 0.64ᵇ | 22.91 ± 0.35ᵇ | 74.31 ± 0.51ᵃ |
| T2        | 25.46 ± 0.58ᵃ | 1.17 ± 0.24ᵃ | 22.43 ± 0.45ᵃ | 22.64 ± 0.64ᵃ | 74.54 ± 0.58ᵃ |
| T3        | 25.34 ± 1.06ᵃ | 1.23 ± 0.29ᵇ | 22.39 ± 0.78ᵃ | 22.39 ± 1.17ᵃ | 74.66 ± 1.06ᵃ |
| T4        | 24.99 ± 0.91ᵃ | 1.09 ± 0.17ᵇ | 22.26 ± 1.01ᵃ | 22.04 ± 1.35ᵃ | 75.00 ± 0.91ᵃ |

Note:
Number which are followed by similar alphabet in the same column are similar during Duncan test of 5% Alphabet a means there is no differences of DM, OM, CP and water of meat. Alphabet b means there is differences CF of meat between control (T0) with T1, T2, T3 and T4.

According to table 3, it can be seen that S. enteridis dose 10¹⁰ CPU (T1) is not affecting the percentage of the meat nutrients compositions since the dose of infection is still low. The low dose of infection is not enough to cause damage to the digestive system so that it was still functioning normally in digesting and metabolizing nutrients and cause the decreasing of CF percentage. The decreasing of CF in treatments T2, T3, and T4, is related to saponin compounds in Z. zerumbet, it can bind the fat from feed in the intestinal. Moreover, it also can bind the cholesterol and bile acids, thus reducing the cholesterol absorption and its biosynthesis in the liver.

4. Conclusions

The addition of Z. zerumbet extract as feed additives to the level of 1 % has not been able to improve the production performance, such as feed consumption, daily weight gain, final weight and normal percentage of carcasses. Besides, it can also decrease the crude fat in the of meat broiler chickens with Salmonelosis.

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References
[1] Guard-Petter J 2001 The Chicken, the egg and Salmonella enteridis Environ. Microbiol. 3(7) 421–30. https://onlinelibrary.wiley.com/doi/epdf/10.1046/j.1462-2920.2001.00213.x
[2] Ariyanti T and Supar 2005 Peranan Salmonella enteritidis pada ayam dan produknya [The role of Salmonella enteritidis in chicken and its product] Wartazoa 15(2) 57–65. [in Bahasa Indonesia] http://medpub.litbang.pertanian.go.id/index.php/wartazoa/article/view/827/836
[3] Ariyanti T and Supar 2008 Antigenisitas dan imunogenisitas salmonella enteritidis: implikasinya dalam diagnosis dan pengembangan vaksin lokal untuk unggas [Antigenecity and immunogenecity of Salmonella enteritidis: Its implication for diagnosis and development of local isolate vaccine for poultry] Wartazoa 18(4) 187–97. [in Bahasa Indonesia] http://medpub.litbang.pertanian.go.id/index.php/wartazoa/article/view/893/902
[4] Thung T Y, Mahyudin N A, Basri D F, Radzi C W J W M, Nakaguchi Y, Nishibuchi M and Radu S 2016 Prevalence and antibiotic resistance of Salmonella enteritidis and Salmonella typhimurium in raw chicken meat at retail markets in Malaysia Poult. Sci. 95(8) 1888–93. https://academic.oup.com/ps/article/95/8/1888/2563779
[5] Sajid A, Kashif N, Kifayat N and Ahmad S 2016 Detection of antibiotic residues in poultry meat J. Pharmacol. Sci. 29(5) 1691–94. https://scholar.google.co.id/scholar?hl=id&as_sdt=0%2C5&q=Detection+of+Antibiotic+Residues+in+Poultry+Meat&btnG=
[6] Abulreesh H H 2012 Salmonellae in the environment Salmonella-distribution, adaptation, control measures and molecular technologies ed B Annous (London: IntechOpen) https://www.researchgate.net/publication/235914938_Salmonellae_in_the_Environment/links/02bfe5141ad4c629d0000000/Salmonellae_in_the_Environment.pdf?origin=publication_detail
[7] Singh C B, Nongallelma Kh, Singh S B, Swapana.N and Singh C D 2011 Zingiber zerumbet Smith – an important medicinal of Zingiberaceae family NeBIO 2(4) 9–13. https://www.researchgate.net/publication/282275034_Zingiber_zerumbet_Smith__an_important_medicinal_plant_of_Zingiberaceae_family
[8] Ganapathy and Nair 2017 Curcuminooids in Zingiber zerumbet rhizome: Bioguided fractionation and chromatographic identification of antimicrobial and antioxidant metabolites J. Herbs Species Med. Plants 23(2) 169–81. https://www.tandfonline.com/doi/abs/10.1080/10496475.2017.1283555
[9] Kader G, Nikkon F, Rashid M A and Yeasmin T 2011 Antimicrobial activities of the rhizome extract of Zingiber zerumbet Linn. Asian Pac. J. Trop. Biomed. 1(5) 409–12 https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3614197/
[10] Dai D N, Tang T D, Chau L T M and Ogunwande I A 2013 Chemical constituents of the root essential oils of Zingiber rubens Roxd and Zingiber zerumbet (L.) Smith Am. J. Plant Sci. 4 7–10. https://scholar.google.co.id/scholar?hl=id&as_sdt=0%2C5&q=Chemical+Constituents+of+the+Root+Essential+Oils+of+Zingiber+Rubens+Roxd%2C+Zingiber+Zerumbet+(L.)+Smith&btnG=
[11] Platel K and Srinivasan K 2004 Digestive stimulant action of spices: A myth or reality Indian J. Med. Res. 119 167–79. https://www.researchgate.net/publication/8489294_Digestive_stimulant_action_of_spices_A_myth_or_reality
[12] Widodo W, Rahayu I D, Sutanto A and Anggraini A D 2017 Penambahan lempuyang dalam pakan ayam Kampung Super yang menggunakan campuran jamu [The addition of lempuyang in Kampung Super chicken feed that uses a mixture of herbs] Seminar Nasional dan Gelar Produk [National Seminar and Product Title] (Malang, Indonesia: Universitas Muhammadiyah Malang) pp 469–73 [in Bahasa Indonesia] http://research-report.unm.ac.id/index.php/research-report/article/view/1256/1474

[13] Irianto A B, Atmomarsono U and Suprijatna E 2014 Pengaruh penambahan tepung jahe merah (Zingiber officinale var. Rubrum) dalam ransum terhadap efesiensi penggunaan protein pada ayam kampung periode pertumbuhan (16–22 minggu) [The effect of added red ginger meal (Zingiber officinale var. Rubrum) in the diet on the Efficiency Protein Utility in Growing Period of Native Chickens Diet (16–22 Weeks)] Animal Agriculture Journal 3(1) 61–9 [in Bahasa Indonesia] https://ejournal3.undip.ac.id/index.php/aaj/article/view/11065

[14] Nasution R A P, Atmomarsono U and Sarengat W 2014 Pengaruh penggunaan tepung daun katuk (Sauropus androgynus) dalam ransum terhadap performa ayam broiler [Influence of katuk (Sauropus androgynus) leaf powder in the diet of broiler performance] Animal Agriculture Journal 3(2) 334–40 [in Bahasa Indonesia] https://ejournal3.undip.ac.id/index.php/aaj/article/view/11489

[15] Gunawan I W G, Bawa I G A G and Sutrisnayanti N L 2008 Isolasi dan identifikasi senyawa terpenoid yang aktif antibakteri pada herba meniran (Phyllanthus niruri Linn.) [Isolation and Identification of terpenoid, antibacterial compounds meniran herb (Phyllanthus niruri Linn.)] Jurnal Kimia 2(1) 31–9 [in Bahasa Indonesia] https://ojs.unud.ac.id/index.php/jchem/article/view/2695

[16] Habibah A. S, Abun and Wiradimadja R 2012 Performan ayam broiler yang diberi ransum mengandung ekstrak kutil jengkol (Pithecellobium jirigina (Jack) Prain) [The performance of broiler chickens given rations contains jengkol skin extract (Pithecellobium jirigina (Jack) Prain)] Fakultas Peternakan. Universitas Padjadjaran, Sumedang [Online] from http://journal.unpad.ac.id/ejournal/article/download/893/939 [Accessed on 20 February 2019]

[17] Chang C J, Tzeng T F, Liou, S, Chang Y and Liu I 2012 Acute and 28-day subchronic oral toxicity of an ethanol extract of Zingiber zerumbet (L.) Smith in rodents Evidence-Based Complementary and Alternative Medicine 2012 1–11 https://www.hindawi.com/journals/ecam/2012/608284/abs/

[18] Manullang J R and Ardhani F 2015 Efektifitas jahe merah (Zingiber officinale Var. Rubrum) sebagai additif pakan dan antimikrobia terhadap pertumbuhan bakteri anaerob dan coliform secara in vivo pada ayam pedaging [The effectiveness of red ginger (Zingiber officinale Var. Rubrum) as feed additive and anti-microbial agent to the in vivo Growth of Anaerob Bacteria and Coliform at Broiler] Jurnal Peternakan Indonesia 17(3) 195–99 [in Bahasa Indonesia] http://jpi.faterna.unand.ac.id/index.php/jpi/article/view/233/196

[19] Darsana I G O, Besung I N K and Mahatmi H. 2012 Potensi daun binahong (Anredera cordifolia (Tenore) Steenis) dalam menghambat pertumbuhan bakteri Escherichia coli secara in vitro [The potential of binahong leaves (Anredera cordifolia (Tenore) Steenis) in inhibiting the growth of Escherichia coli bacteria in vitro] Indonesia Medicus Veterinus 1(3) 337–51 [in Bahasa Indonesia] https://ojs.unud.ac.id/index.php/imv/article/view/1879

[20] Ketaren P P 2010 Kebutuhan gizi ternak unggas di Indonesia [Nutritional needs of poultry in Indonesia] Warta佐a 20(4) 172–80.[in Bahasa Indonesia] https://anzdoc.com/kebutuhan-gizi-ternak-unggas-di-indonesia.html