

Effectiveness of Fermented Star Fruit (*Averrhoa bilimbi* L.) Filtrate Acidifier on Growth Performance, Blood Hematology Profile, and Percentage Carcass of Heat-Stressed KUB Chickens

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Abstract | Heat stress is a major constraint in poultry production in tropical regions and can reduce the growth performance and physiological health of Balitbangtan Superior Village (KUB) chickens. Most research on natural acidifiers to date has focused on commercial broilers, so their use in KUB chickens has not been widely discussed in the literature. This study aims to evaluate the effectiveness of fermented belimbing wuluh fruit (*Averrhoa bilimbi* L.) filtrate (FABF) as a natural acidifier in drinking water on the production performance, haematological profile and percentage carcass of heat-stressed KUB chickens. A total of 320 KUB chickens DOC were used in randomized complete factorial design with two factors, FABF concentration (0%, 0.5%, 1.0%, and 1.5%) and cage temperature (32°C, 34 °C 36 °C and 38°C). The parameters observed were feed consumption, drinking water consumption, body weight gain, feed conversion, blood hematology profile and percentage carcass. Data were analyzed using Analysis of Variance (ANOVA) and Duncan Multiple Range Test (DMRT) further tests. The results showed that the administration of fermented star fruit filtrate had a significant effect ($p < 0.05$) in increasing feed intake, body weight gain, live weight, carcass percentage, and commercial meat cut percentage, as well as improving the blood hematology profile of KUB chickens. The optimal treatment was obtained at a concentration of 1 % with a cage temperature of 32°C. Based on these findings, FABF has the potential to be used as a natural acidifier and an alternative to synthetic antibiotics, thereby supporting poultry productivity in conditions of tropical heat stress.

Keywords | KUB chickens, Star fruit, *Acidifier*, Growth performance, Blood hematology, Percentage of carcass

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INTRODUCTION

Global climate change and increasing ambient temperatures have a significant impact on livestock performance, specifically poultry. Heat stress is a physiological condition that occurs when chickens are unable to compensate for high environmental temperatures, leading to decreased body weight, increased feed conversion

ratio, and mortality (Tamzil, 2014). Balitbangtan Superior Village (KUB) chickens are local strains developed to support animal protein self-sufficiency. KUB is also at risk of reduced performance when kept in extreme heat conditions (Priyanti *et al.*, 2016).

The negative effects of heat stress can be seen in reduced feed intake, body weight growth, and feed conversion

efficiency, as well as disruption of physiological parameters such as decreased hemoglobin, hematocrit, and erythrocyte levels in the blood (Sugito and Mira, 2009). A recent method applied to address these issues is nutritional intervention by adding natural additive such as acidifier. In this context, acidifier is known to lower the pH of gastrointestinal tract, suppress the growth of pathogenic bacteria, and improve the balance of the gut microbiota, contributing to improved digestibility and immune status (Adil *et al.*, 2020). Acidifier is categorized into synthetic and natural forms. The use of natural acidifier is an environmentally friendly alternative while supporting the ban on the use of antibiotic growth promoters (AGPs) in various countries. The growing concern over antimicrobial resistance has increased the interest in natural acidifiers as alternatives to antibiotic growth promoters (AGPs) in poultry diets. Most previous studies have focused on commercial acidifiers such as vinegar, citrus extracts, or synthetic organic acids, particularly in fast-growing broiler strains. The fermentation process can increase the availability of bioactive compounds and enrich probiotic microorganisms, providing a synergistic effect in suppressing the effects of stress and improving livestock performance (Firmansyah *et al.*, 2022). Among various natural acidifiers, *Averrhoa bilimbi* (star fruit) is a promising candidate due to its unique bioactive composition. This fruit contains high levels of organic acids, including oxalic, citric, acetic, formic, fumaric, malic, succinic, tartaric, axaloacetic, quinic and shikimic acids, which are effective in lowering gastrointestinal pH, thereby enhancing nutrient utilization and suppressing pathogenic microorganisms (Muhtahdi *et al.*, 2011). In addition, *A. bilimbi* is rich in secondary metabolites such as flavonoids, phenolics and saponins that exhibit antimicrobial and antioxidant properties, potentially supporting gut health and resilience under heat stress conditions. Another advantage of this fruit is its abundance in tropical regions, making it a locally available, economical, and sustainable resource compared to many commercial acidifiers. These characteristics provide a strong rationale for evaluating the effectiveness of fermented *A. bilimbi* filtrate as a natural acidifier to improve growth performance, hematological profile, and carcass yield of KUB chickens exposed to heat stress.

Heat stress is one of the major limiting factors in poultry production in tropical environments, as ambient temperatures above 32°C can disrupt physiological functions, reduce feed intake, and impair growth performance. In this study, a temperature range of 32–38°C was selected, since preliminary trials indicated that exposure to 40°C induces severe heat stress and increases mortality risk, which is not acceptable under animal welfare and ethical research standards (Lara and Rostagno,

2013). Moreover, the Indonesian Ministry of Agriculture has banned the use of antibiotic growth promoters (AGP) through Regulation of the Minister of Agriculture No. 14 of 2017, highlighting the need for natural alternatives such as locally sourced acidifiers. Fermented starfruit (*Averrhoa bilimbi* L.) filtrate contains natural organic acids with potential as an acidifier and has not been widely studied for its use in local KUB chickens. While numerous alternatives to antibiotic growth promoters (AGPs) including probiotics, prebiotics, and phytogenics have been extensively investigated and implemented in poultry production, acidifiers present a distinctive mode of action that is particularly advantageous under heat stress conditions, by reducing gastrointestinal pH, acidifiers inhibit the proliferation of harmful microorganisms such as *E. coli* and *Salmonella*, while at the same time promoting digestive enzyme function and improving nutrient absorption. These effects are especially important during periods of heat stress, when feed intake and nutrient utilization are typically compromised. Furthermore, natural acidifiers sourced from tropical plants, such as *Averrhoa bilimbi* (star fruit), contribute additional value due to their abundance of organic acids and bioactive constituents with antimicrobial and antioxidant activities. This combination of acidifying action and bioactive protection positions acidifiers not only as promising substitutes for AGPs but also as particularly effective agents in alleviating the physiological and metabolic challenges associated with heat stress in poultry.

Previous reports have demonstrated that synthetic acidifiers, such as citric and formic acid or their combinations, can improve nutrient digestibility and promote body weight gain in broiler chickens (Huyghebaert *et al.*, 2011; Adil *et al.*, 2020). However, the scientific use of locally sourced fermented natural acidifiers, such as *Averrhoa bilimbi* filtrate for native KUB chickens, remains poorly documented, highlighting a specific area that requires further investigation. Research on the utilization of natural acidifier sources from local tropical ingredients such as star fruit in the form of fermented filtrate are still very limited. Furthermore, very few analyses have tested the effects of using natural acidifier on local chickens such as KUB with different physiological characteristics from broilers.

Kampung Unggul Balitbangtan (KUB) chickens are an improved native Indonesian breed with slower growth rates and lower feed efficiency compared to commercial broilers, making them suitable for studies on nutrient optimization. They possess distinct hematological and immune characteristics, which may lead to different responses to dietary interventions such as fermented star fruit filtrate (FABF). Although more tolerant to tropical heat than broilers, KUB chickens still experience reduced performance under prolonged heat stress. Therefore,

evaluating natural acidifiers like FABF in KUB chickens has both scientific relevance and practical value for poultry production in tropical smallholder systems. Therefore, this research aims to evaluate the effectiveness of fermented star fruit filtrate as acidifier in drinking water on the production performance, blood hematology profile and percentage carcass of heat-stressed KUB chickens. The results contribute to the development of natural feed technology adaptive to climate change and support a sustainable local poultry farming system.

MATERIALS AND METHODS

This research was conducted from September to November 2024 at the Poultry Laboratory and Animal Physiology and Health Laboratory, Faculty of Animal Husbandry Halu Oleo University. The equipment included brooder and treatment cages, feed and drinking water containers, incandescent lamps, scales, temperature control devices (blower, hygrometer, thermostat), a Philips blender with a capacity of 1.5 l, Whatman paper, a vacuum pump, 500 ml and 1 l Erlenmeyer tubes, digital scales, spoit, blood sampling equipment (syringe, EDTA vacutainer), and laboratory equipment such as Hematology autoanalyzer, incubator, Erlenmeyer tubes, pH meter, test tubes, drop pipettes, micropipettes, Petri dishes, gloves, and camera. A total of 320 KUB chickens DOC from PT Jatinom Unggas Perkasa were used in this research. Other materials included rice husk, ND vaccine, sugar water, commercial feed (BP 11, Table 1), basal feed for the grower phase, and star fruit obtained from a garden in Kendari City.

PREPARATION OF FERMENTED *AVERRHOA BILIMBI* FILTRATE (FABF)

Fresh *Averrhoa bilimbi* fruits were sourced from the Mandonga traditional market (Kendari, Indonesia). The fruits were carefully rinsed under running water, sliced into pieces of approximately 2 cm, and blended using a 1.5 l blender until homogenized. The resulting mixture was filtered through Whatman No. 42 paper to obtain a clear filtrate. The initial pH of the filtrate was recorded at 4.5 ± 0.1 using a digital pH meter (Hanna Instruments). To initiate fermentation, the filtrate was inoculated with 1% (v/v) *Lactobacillus* sp. and placed into sterile vacuum-sealed jars to maintain anaerobic conditions. Fermentation was performed at 30 ± 2 °C for 4 days, after which the pH dropped to 3.2 ± 0.1 , indicating successful lactic acid fermentation. The fermented filtrate was subsequently diluted to prepare three treatment concentrations for drinking water supplementation: 0.5% (pH 3.8 ± 0.1), 1.0% (pH 3.6 ± 0.1), and 1.5% (pH 3.4 ± 0.1). All equipment was sterilized with 70% ethanol, and the FABF solutions were freshly prepared with pH verified before administration to chickens (Figure 1).

ETHICAL APPROVAL

The ethical committee of Halu Oleo University Kendari, Southeast Sulawesi, Indonesia approved this study with reference number 248/UN29,20/EC/2024, dated July 10th, 2024, and followed standard Animal Welfare Guidelines. Chickens maintained at 38 ± 1 °C were observed at 30 minute intervals to detect potential signs of heat stress. Corrective measures, including mist spraying or enhanced ventilation, were administered only when severe stress indicators appeared and accounted for less than 5% of the total monitoring period. These measures were minimal, temporary, and did not affect the overall room temperature. To ensure treatment consistency, temperature and humidity in each group were continuously monitored with a digital data logger, confirming that no significant microclimate variations influenced the experimental conditions.

EXPERIMENTAL DESIGN

This study used a 4×4 factorial completely randomized design (CRD), with fermented starfruit filtrate concentrations (0%, 0.5%, 1.0%, 1.5%) and cage temperatures (32°C, 34°C, 36°C, 38°C). A total of 320 DOC KUB chickens were divided into 16 treatment combinations, each with 4 replications, so there are 64 experimental units. Four replications were considered adequate based on the homogeneity of the initial population (CV <10%) and the minimum requirements for ANOVA analysis for factorial designs (Steel and Torrie, 1980). A post-hoc power analysis was conducted based on the variability, and the results indicated that the sample size used in this study provided a statistical power greater than 80%, confirming that the number of birds per treatment was sufficient to detect significant differences. The treatment lasted for 8 weeks. Parameters observed included feed consumption, drinking water consumption, body weight gain, feed conversion, carcass percentage, commercial meat cut percentage, and hematological profile.

OBSERVED VARIABLES

Observed production performance parameters included feed consumption, drinking water consumption, body weight gain (PBB), feed conversion (FCR), and hematological profile (erythrocyte count, leucocytes, hemoglobin level, hematocrit value, lymphocytes, and erythrocyte index (Mean Corpuscular Volume (MCV))). Feed consumption (g/head) (Rasyaf, 1994):

$$\text{Feed Consumption} = \frac{\text{Feed given during the treatment} - \text{Feed residue}}{\text{Number of chickens}} \times 100\%$$

Drinking water consumption (mL/head). Drinking water consumption was measured from the amount given minus the daily remnant, specifically during the administration of fermented star fruit filtrate from 5 to 8 weeks of age.

Body Weight Gain (g/head) (Rasyaf, 1994).

$$\text{Body weight gain} = \frac{BB \text{ The end} - BB \text{ The beginning}}{\text{Maintenance time}}$$

$$\text{Feed conversion} = \frac{\text{Feed consumption amount}}{\text{Body weight gain}}$$

Feed Conversion (Rasyaf, 1994).

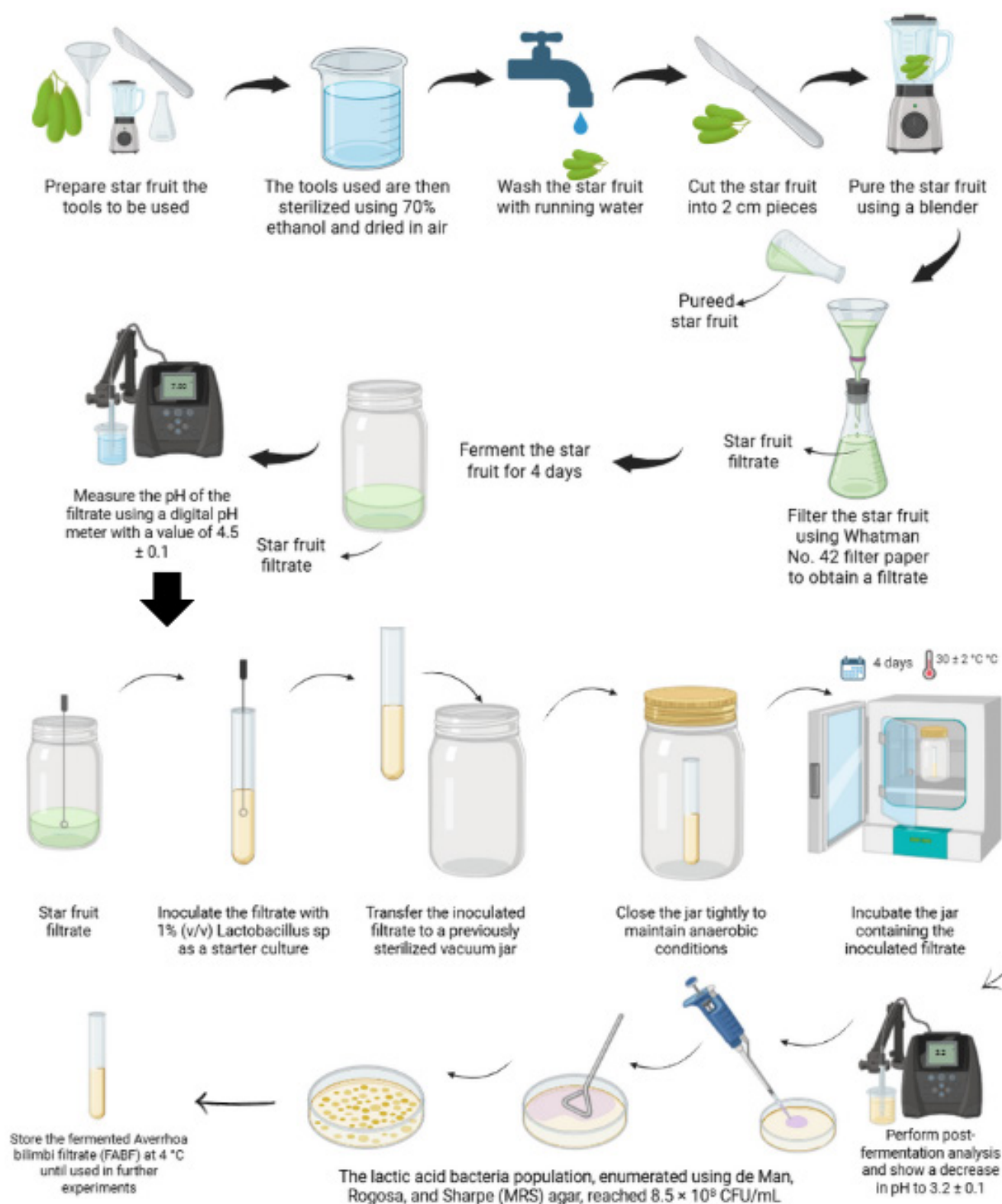


Figure 1: Preparation of fermented star fruit filtrate according to a modified method by Pratama *et al.* (2021) resulting in FABF.

BLOOD SAMPLING

Blood sampling was conducted when chickens were 60 days old by taking 1 randomly in each experimental plot. Blood collection was achieved on the brachial vein located on the ventral part of the wing. Samples of 2 mL taken from each chicken were put into a tube containing EDTA anticoagulant and shaken gently (Napirah, 2013). The samples were stored in a coolbox to be taken to the laboratory for analysis. The principle was to accurately count and measure based on the volume and number of cells in the blood (DeNicola, 2011). Hematological profile included erythrocyte count, leucocytes, hemoglobin, lymphocytes, hematocrit, and erythrocyte index (MCV).

CARCASS PERCENTAGE AND COMMERCIAL CARCASS PARTS PERCENTAGE

After the feather removal process, the giblets, head, neck, and legs of chickens were separated or cut to obtain the whole carcass. The carcass was weighed, and separated into the thigh, wing, back, and breast sections. Subsequently, each of these carcass sections was weighed before calculating the percentage of the section. Live weight could be obtained by weighing broiler chickens fasted before slaughter. The carcass percentage was obtained by comparing the carcass and live weight multiplied by 100% (Soeparno, 2009).

$$\text{Percentage of carcass} = \frac{\text{Weight of carcass}}{\text{Live weight}} \times 100\%$$

$$\text{Percentage of carcass parts} = \frac{\text{Weight of carcass parts}}{\text{Weight of carcass}} \times 100\%$$

STATISTIC ANALYSIS

The data obtained was analyzed using Analysis of Variance (ANOVA). Further tests were carried out using Duncan Multiple Range Test (DMRT) through SPSS program when the treatment was significantly different (Steel and Torrie, 1993).

RESULTS AND DISCUSSION

PERFORMANCE OF KUB CHICKENS

ANOVA showed that the combination of acidifier from fermented star fruit filtrate and cage temperature after heat stress had a significant effect on the feed consumption of KUB chickens ($p < 0.05$). Cage temperature and filtrate concentration variation separately had a significant effect. Feed consumption of KUB chickens after heat stress ranged from 2700 to 4471.8 g/bird, with the best results obtained in the combined treatment of 1% filtrate and 32°C cage temperature (K3T1). This is higher than the results of previous research of 3323.65 g/head (Wiradimadja *et al.*, 2015). A decrease in feed consumption occurred after chickens experienced heat stress at 38°C due to metabolic

disorders and stress. A significant increase in consumption occurred at week 7 after 3 weeks of fermented star fruit filtrate was administered through drinking water. Active compounds such as antioxidant, vitamin C, and anti-stress compounds helped chickens recover from heat stress. Star fruit contains various compounds acting as antimicrobial, immunomodulator, antioxidant, and acidifier (Pratama *et al.*, 2021). However, the effectiveness of filtrate decreased after week 7 due to an insignificant impact on feed consumption by week 8.

This research showed that there was a significant interaction ($p < 0.05$) between the provision of fermented star fruit filtrate acidifier and variations in cage temperature after heat stress on drinking water consumption of KUB chickens. In addition, cage temperature treatment and acidifier type had a significant effect on drinking water consumption. The range of drinking water consumption of KUB chickens ranged from 152.9 to 177.1 ml/head, lower than (Sari, 2023) with a range of 386–417.2 ml/head. The highest and lowest consumptions were obtained in the treatment combinations K2T1 (0.5% filtrate and 32°C cage) and K2T2 (0.5% filtrate and 34°C), respectively. In this study, FABF was administered at its natural post-fermentation pH (3.2 ± 0.1), which likely contributed to reduced water intake due to lower palatability. The decrease in drinking water consumption was caused by the low pH of the water (3.67–3.74) due to citric acid as acidifier given ad libitum. This was in line with Chowdhury *et al.* (2009) since the optimal pH of drinking water for chickens ranged from 5 to 8. Excessive administration of organic acid could reduce water and feed consumption as well as chicken growth (Ross, 2018). Meanwhile, the extremely acidic pH value in this research was not tolerated by KUB chickens, potentially reducing water and ration consumption. Chickens with low environmental temperatures consumed lower feed and water (Jahejo *et al.*, 2016).

Table 1: Nutrient content of feed BP 11.

Nutritional composition	Nutritional content
Ash	7%
Crude protein (CP)	21%
Eter extract (EE)	3%
Crude fiber (CF)	5%
Calcium (Ca)	1.1%
Phosphorus (P)	0.9%
Metabolic energy (ME)	3000 kcal/Kg

Source: PT. Charoen Pokphand Indonesia Tbk. 2023.

ANOVA results showed that there was a significant interaction ($p < 0.05$) between the treatment of fermented star fruit filtrate and cage temperature on the body weight gain of KUB chickens. Furthermore, the factors had a significant

effect ($p < 0.05$), and the range of body weight gain of KUB was between 302–463 g/head (Table 2). This was in line with the results of previous research by Urfa *et al.* (2017) which reported 451.3–512.0 g/head. The best treatment combination was found in K3T1 at 1% filtrate with 32°C cage temperature, resulting in the highest weight gain of 463 g/head. The greatest improvement in body weight gain occurred with 1.0% FABF supplementation, while increasing the dose to 1.5% failed to provide additional benefits, likely due to diminished palatability and a possible saturation effect. This emphasizes the need to carefully determine the optimal FABF level to maximize its positive impact while maintaining adequate feed and water intake in heat-stressed KUB chickens. The increase in body weight of rearing was influenced by the organic acid content in star fruit filtrate. Bioactive components in star fruit, such as antimicrobial, antioxidant, immunomodulator, and acidifier, also support the improvement of chicken growth and health. This result was in line with Wahyuni *et al.* (2020) and Pratama *et al.* (2021) which showed the potential of star fruit fermentation filtrate as a natural alternative to AGP (Antibiotic Growth Promoter) in improving chicken performance.

Interestingly, the 1.5% FABF treatment did not always result in a higher body weight gain compared to the 1.0% dose, although both were statistically similar. This result may be attributed to the saturation effect of acidifiers, where an excessive concentration of organic acids can reduce palatability or cause suboptimal gastrointestinal pH conditions, which slightly decrease feed intake despite better physiological status. This observation is consistent with Huyghebaert *et al.* (2011), who reported that acidifier doses exceeding the optimal point may negatively affect growth performance. Thus 1.0% concentration appears more effective for improving growth, whereas 1.5% is more optimal for enhancing hematological parameters and carcass traits. This indicates that the response of chickens to acidifiers are not always linear and that the optimal dose depends on the specific performance variable measured. Although the fermented filtrate supported physiological resilience, the increased feed intake at 1.5% resulted in greater energy consumption without a proportional increase in weight gain, leading to a higher FCR, especially during the post-heat-stress recovery phase.

Table 2: Mean feed consumption, drinking water consumption, body weight gain and ration conversion of KUB chickens fed with different concentrations of fermented star fruit filtrate (*Averrhoa bilimbi* L) and cage temperature after heat stress.

Parameters	Filtrate concentration (%)	Cage temperature (°C)				Average
		T1	T2	T3	T4	
Feed consumption (g/head)	K1	4167.6±52.75 ⁱ	4621.8±92.68 ^j	3195±26.07 ^{ef}	2900±443.47 ^{cd}	3721.1±195.08 ^c
	K2	4448.4±74.05 ^j	3294.4±112.25 ^f	2934.4±17.59 ^a	2704±7.00 ^b	3645.3±49.41 ^a
	K3	4471.8±89.44 ⁱ	3927.8±266.85 ^h	2799.2±48.95 ^{bc}	2895±23.72 ^{cd}	3523.5±109.80 ^b
	K4	4464.0±50.42 ^j	3712.2±29.44 ^g	3053.4±20.39 ^{de}	2700±38.24 ^b	3482.4±12.81 ^b
	Average	4387.0±195.08 ^d	3889.1±183.14 ^c	2364.0±209.95 ^a	2494.5±175.64 ^b	
Water consumption (ml/head)	K1	148.4±5.08 ^a	163.6±4.04	163.2±2.16 ^{cd}	167.4±0.44 ^f	160.6±2.05 ^a
	K2	177.1±2.83 ^g	152.9±2.23 ^b	163.1±1.63 ^{cd}	166.8±1.28 ^{ef}	164.9±0.68 ^c
	K3	153.4±1.03 ^b	162.2±1.29 ^c	166.6±0.70 ^{def}	169.0±0.66 ^f	162.8±4.37 ^b
	K4	154.2±4.99 ^b	167.1±0.68 ^{ef}	156.0±3.47 ^b	170.1±1.42 ^f	161.8±3.10 ^{ab}
	Average	158.3±55.28 ^a	161.4±57.29 ^b	162.2±58.90 ^b	168.3±62.34 ^c	
Body weight gain (g/head)	K1	432.6±48.35 ^c	378.6±25.35 ^{bc}	368.6±17.97 ^{bc}	310.0±21.41 ^a	372.4±13.72 ^{ab}
	K2	435.6±15.76 ^c	398.8±18.01 ^{cd}	397.0±14.68 ^c	430.6±13.85 ^{de}	415.5±1.80 ^c
	K3	463.0±29.97 ^c	398.6±16.46 ^{cd}	305.6±8.38 ^a	302.6±11.89 ^a	367.4±9.46 ^a
	K4	347.4±32.78 ^b	456.8±30.29 ^c	391.0±28.52 ^c	354.4±21.14 ^b	387.4±5.01 ^b
	Average	419.6±13.72 ^c	408.2±4.95 ^c	365.5±3.85 ^b	349.4±4.92 ^a	
Feed conversion ratio	K1	9.73±0.88 ^{ef}	12.25±0.90 ^g	8.60±0.42 ^{cd}	9.91±0.37 ^{ef}	10.12±0.29 ^d
	K2	10.22±0.39 ^f	8.28±0.34 ^{bc}	7.44±0.24 ^{ab}	6.81±0.21 ^a	8.19±0.08 ^a
	K3	9.90±0.53 ^{ef}	9.69±0.87 ^{ef}	9.16±0.23 ^{de}	9.46±0.49 ^{de}	9.56±0.27 ^c
	K4	12.75±1.34 ^g	8.14±0.50 ^{bc}	7.64±0.59 ^{ab}	7.65±0.45 ^{ab}	9.05±0.48 ^b
	Average	10.65±0.29 ^c	9.59±0.28 ^b	8.21±0.07 ^a	8.46±0.06 ^a	

Different superscript letters in the same row and column indicate that the treatment has a significant effect ($p < 0.05$). K1 (0% filtrate concentration), K2 (0.5% filtrate concentration), K3 (1.0% filtrate concentration), K4 (1.5% filtrate concentration). T1 (cage temperature 32°C), T2 (cage temperature 34°C), T3 (cage temperature 36°C), T4 (cage temperature 38°C)

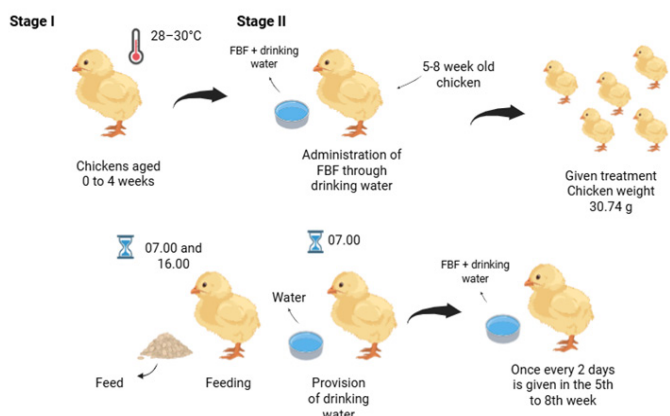


Figure 2: Research procedure for administering star fruit fermentation filtrate to drinking water for KUB chickens aged 5 to 8 weeks.

The provision of fermented filtrate of star fruit and cage temperature after heat stress interacted significantly ($p < 0.05$) on the ration conversion value of KUB chickens. Despite the significant effect, the range of ration conversion obtained (7.44-12.75) was higher than the results of Wicaksono *et al.* (2013) at 5.0-5.5, indicating lower feed efficiency. Although the fermented filtrate improved carcass traits and blood parameters, it did not proportionally lower the FCR, likely because higher intake at the 1.5% level increased energy consumption without a matching improvement in body weight gain. This indicates that while the filtrate enhanced physiological resilience, its efficiency in feed conversion may be more dose-dependent and limited under post-stress recovery conditions. The lower efficiency led to high ration consumption due to the effect of fermentation filtrate slowing down intestinal peristalsis. In addition, the condition of KUB chickens in the recovery phase allowed the nutrients to be used for recovery, increasing the feed conversion value. However, fermented star fruit filtrate showed potential in aiding digestive tract recovery and nutrient absorption through organic acid content. The effectiveness of filtrate appeared to be limited up to week 6 of rearing. Organic acid could improve gastrointestinal function and growth performance of chickens (Sugiharto, 2021).

BLOOD HEMATOLOGY PROFILE OF KUB CHICKENS

The provision of fermented star fruit filtrate in drinking water with variations in cage temperature had a significant effect ($p < 0.05$) on the number of blood erythrocytes of KUB chickens, with a range of $3.51-3.95 \times 10^6/\text{mm}^3$ (Figure 3). This value was higher than the normal standard of $2.00-3.20 \times 10^6/\text{mm}^3$ (Smith and Soesanto, 1988). The increase in erythrocytes was due to the effect of extreme temperature and increased filtrate concentration rich in essential nutrients such as organic acid, vitamin, and mineral. The best treatment was obtained in the combination of K3T1 (1% filtrate, 32 °C temperature)

with the highest and lowest number of erythrocytes at $3.95 \times 10^6/\text{mm}^3$ and $3.51 \times 10^6/\text{mm}^3$, respectively. The nutritional content of filtrate supported the formation of red blood cells through the fulfillment of protein and vitamin requirements. The results showed that the provision of fermented star fruit filtrate in drinking water with variations in cage temperature had a significant effect ($p < 0.05$) on the blood leucocyte count of KUB chickens, with a range of $5.44-8.52 \times 10^3/\text{mm}^3$. This value was lower than the normal standard of $16-40 \times 10^3/\text{mm}^3$ influenced by heat stress and the physiological conditions of chickens (Smith and Soesanto, 1988). The total leukocyte count in this study ($5.44-8.52 \times 10^3/\text{mm}^3$) was lower than the normal range ($16-40 \times 10^3/\text{mm}^3$). However, some literature indicates that local chickens such as KUB tend to have physiologically lower leukocyte counts than broiler chickens (Rehman *et al.*, 2016). More recent reports indicate that Indonesian native chickens generally present lower baseline leukocyte counts than commercial strains, which is linked to their distinct genetics and better adaptation to tropical environments.

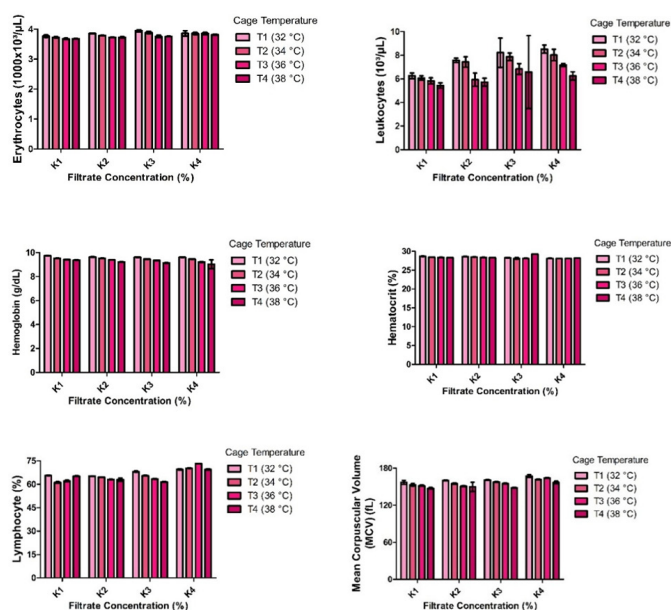


Figure 3: The average blood erythrocyte, leucocyte, hemoglobin, hematocrit, lymphocyte and erythrocyte index (MCV) counts of KUB chickens treated with different concentrations of fermented star fruit filtrate (*Averrhoa bilimbi* L) and different cage temperatures after heat seclusion. K1 (0% filtrate concentration), K2 (0.5% filtrate concentration), K3 (1.0% filtrate concentration), K4 (1.5% filtrate concentration), T1 (cage temperature 32°C), T2 (cage temperature 34°C), T3 (cage temperature 36°C), T4 (cage temperature 38°C).

These differences reflect genetic adaptation rather than pathology, indicating that the observed counts fall within the normal physiological range. Furthermore, the stability of leukocyte levels across treatments suggests that FABF

supplementation did not impair immune function under heat stress. Furthermore, the consistent leukocyte levels across treatments demonstrate that FABF supplementation preserved hematological balance under heat stress without causing immunosuppression. In addition to genetic factors, exposure to high temperatures can also reduce immune activity and cause partial immunosuppression through the release of stress hormones (glucocorticoids), which suppress white blood cell proliferation. However, this reference was derived mainly from commercial broiler chickens and may not accurately reflect indigenous breeds such as KUB. The best treatment was recorded in the combination of K1T1 (0% filtrate, 32°C) with the highest leukocyte count of $8.52 \times 10^3/\text{mm}^3$, while the lowest was K1T4 (0% filtrate, 38°C). The increase in leukocytes reflected the immune response of the body to stress and infection and was influenced by temperature, nutrition, and physiological conditions (Ganong, 1995; Soeharsono *et al.*, 2010). The best treatment was recorded in the combination of K1T1 (0% filtrate, 32°C) with the highest leukocyte count of $8.52 \times 10^3/\text{mm}^3$, while the lowest was K1T4 (0% filtrate, 38 °C). The increase in leukocytes reflected the immune response of the body to stress and infection and was influenced by temperature, nutrition, and physiological conditions (Ganong, 1995; Soeharsono *et al.*, 2010).

The analysis of variance reported that the provision of fermented star fruit filtrate in drinking water with variations in cage temperature had a significant effect ($P < 0.05$) on the blood hemoglobin levels of KUB chickens, with a range of 9.03-9.74 g/dL. However, this value was within the normal range of 7.3-10.9 g/dl (Smith and Soesanto, 1988). Treatments K1T1 (0% filtrate, 32°C) and K4T4 (1.5% filtrate, 38°C) produced the highest and lowest hemoglobin levels of 9.74 g/dl and 9.03 g/dl, respectively. Optimal cage temperature played an important role in supporting hemoglobin levels, while fermented starfruit filtrate increased the efficiency of protein metabolism. High hemoglobin reflected the ability to optimally transport oxygen to tissues and carbon dioxide (CO_2), supporting the overall physiological function of the chicken (Cunningham, 2000).

The provision of fermented star fruit filtrate in drinking water with variations in cage temperature had a significant effect ($p < 0.05$) on hematocrit value (PCV) of KUB chicken blood, with a range of 28.07-28.69% in 23-35% (Samour, 2015). The highest and lowest values were obtained in the combination of K1T1 (0% filtrate, 32°C) and K4T2 (1.5% filtrate, 34°C), respectively. The decrease in PCV at high filtrate and extreme temperatures was related to heat stress and decreased nutrient utilization. The content of antioxidant, organic acid, and mineral in star fruit

filtrate could help maintain the stability of erythrocytes and improve red blood cell formation (Zhang *et al.*, 2017). A normal hematocrit showed a good health condition of chickens since the variable was directly proportional to the number and size of erythrocytes formed. Increased hematocrit was caused by the total number of erythrocytes which described the ratio between red blood cells and plasma (Frandsen, 1992).

The results showed that the provision of fermented star fruit filtrate in drinking water with variations in cage temperature had a significant effect ($p < 0.05$) on the percentage of blood lymphocytes of KUB chickens, with a range of 61.2-73.2% in normal limits of 24-84% (Smith and Soesanto, 1988). Duncan test results reported that the treatment combination K4T3 (1.5% filtrate, 36°C) and K3T4 (1% filtrate, 38°C) showed the highest and lowest percentage of blood lymphocytes at 73.2% and 61.2%, respectively. Increasing filtrate concentration to 1.5% tended to improve lymphocytes, while extreme cage temperatures reduced the values. The active ingredients in star fruit such as organic acid, antioxidants, vitamins, and minerals maintain the stability of the immune system, including lymphocyte production. This effect showed the potential of star fruit filtrate as a natural agent to increase the immune system of chickens (Escala *et al.*, 2015).

The provision of fermented star fruit filtrate in drinking water with variations in cage temperature had a significant effect ($p < 0.05$) on MCV level of KUB chicken blood, with a range of 147.40-167.40 fl, exceeding the normal limit of 90-140 fl (Jain. N.C, 1993) dan 104 – 135 fl (Samour, 2015). Duncan test results showed that the treatment combination K1T1 (0% filtrate, 32°C) and K1T4 (0% filtrate, 38°C) reported the highest and lowest blood MCV levels of 167.40 fl and 147.40 fl, respectively. Although the MCV values observed in this study (147–167 fl) exceeded the normal limit of (90–140 fl), this elevation was not accompanied by a decrease in hemoglobin, PCV, or total erythrocyte counts. This suggests a physiologically compensated macrocytosis rather than a pathological macrocytic anemia. Previous reports have noted that native or slow-growing chicken breeds often show higher MCV values compared to fast-growing broilers due to natural differences in erythrocyte morphology (Rehman *et al.*, 2016). Furthermore, mild macrocytosis may arise as a compensatory response during recovery from heat stress, when erythrocytes undergo adaptive enlargement without functional impairment. The increase in MCV is likely due to the crude fiber content of the feed (5%), which can affect nutrient absorption and red blood cell formation, thereby affecting MCV. Hence, the elevated MCV observed likely reflects both an adaptive response to heat stress and the influence of dietary fiber, rather than a

pathological condition. High MCV values were associated with adequate nutrition from feed and optimal ambient temperature, which supported metabolic processes and oxygen distribution. Conversely, extreme cage temperatures decreased MCV and potentially compromised the physiological status of chickens. MCV was influenced by feed, crude fiber, and environmental conditions (Rini *et al.*, 2013).

LIVE WEIGHT, CARCASS PERCENTAGE, AND COMMERCIAL CARCASS PARTS PERCENTAGE OF KUB CHICKENS

ANOVA results showed that the administration of fermented star fruit filtrate at different cage temperatures reported a significant interaction effect ($p < 0.05$) on the slaughter weight of KUB chickens with a live weight range of 777–970.60 g/head (Figure 4). According to Sigaha *et al.* (2019), the live weight of super free-range chickens using conventional feed was 850.75 g/head. used as a reference to compare the performance of local chickens under standard feed conditions This differed from the results reported by (Lorenza and Rita, 2024), where the live weight of KUB chickens was 1153.85–1214.05 g/head. Star fruit contains oxalic acid, which functions as an antimicrobial agent to suppress pathogenic microorganisms, improve nutrient absorption, and support the increased live weight of KUB chickens (Jamilah *et al.*, 2013).

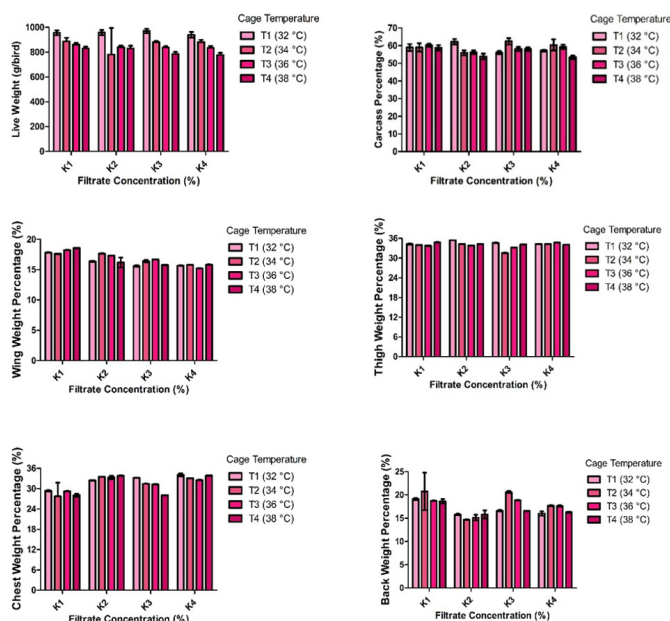


Figure 4: The average live weight, carcass percentage, and commercial carcass cuts percentage of KUB chickens treated with different concentrations of fermented star fruit filtrate (*Averrhoa bilimbi* L) filtrate and different cage temperatures after heat isolation. K1 (0% filtrate concentration), K2 (0.5% filtrate concentration), K3 (1.0% filtrate concentration), K4 (1.5% filtrate concentration), T1 (cage temperature 32°C), T2 (cage temperature 34°C), T3 (cage temperature 36°C), T4 (cage temperature 38°C).

The percentage range of KUB chicken carcasses fed with fermented star fruit filtrate at different cage temperatures was 53.44–62.42%. The results were consistent with Lorenza and Reta (2024) at a percentage range of 62.32–67.87%. Fermented star fruit filtrate acted as acidifier due to the high organic acid content, lowering the pH of the digestive tract and supporting digestive function. This had a positive impact on increasing weight and carcass percentage. Additionally, the natural acid content had the potential to replace antibiotics by inhibiting pathogenic bacteria and stabilizing the gut microbiota of poultry (Gauthier, 2002).

The administration of fermented star fruit filtrate at different cage temperatures showed different interaction effects ($p < 0.05$) on the percentage of breast weight in KUB chickens. The range of breast weight percentages was 28.02–34.05%. The results were higher than Silitonga *et al.* (2024), where the percentage of breast weight in KUB chickens was 26.53–27.65%. Fermentation filtrate of star fruit had a higher natural acid content for improving feed digestibility and increasing the amount of feed directly. Breast muscle was highly responsive to feed factors (Aryani *et al.*, 2019). The observed increase in breast meat yield with FABF supplementation can be explained through specific physiological pathways. As an acidifier, FABF lowers gastrointestinal pH, which enhances enzyme activity and amino acid absorption is essential for muscle protein synthesis. Since the pectoralis major has high metabolic activity and protein turnover, it benefits most from improved amino acid availability. Moreover, the organic acids and bioactive compounds in *Averrhoa bilimbi* provide antimicrobial and antioxidant effects that reduce oxidative stress and promote efficient nutrient partitioning toward lean tissue growth.

The concentration of fermented star fruit filtrate had a significant effect ($P < 0.05$) on thigh weight percentage. However, different cage temperatures did not affect thigh weight percentage ($P > 0.05$), ranging from 31.55 to 35.39%. The results were higher than Qadar *et al.* (2022), where the percentage of thigh weight was 30.84–32.86% due to fermented star fruit. This enhanced palatability and digestibility, allowing food to be directly absorbed and used by livestock to increase thigh meat percentage.

ANOVA reported that the administration of fermented star fruit filtrate at different cage temperatures showed different interaction effects ($P < 0.05$) on wing weight percentage of 15.21–17.83%. The results were higher than Qadar *et al.* (2022), where wing weight percentage was 8.12–9.04%. This was due to the effect of a housing temperature of 32°C on increased feed intake in the normal range for poultry. According to Kurniawan *et al.* (2012), bone tissue growth and development were influenced by

genetic factors, feed, and hormonal functions.

The administration of fermented star fruit filtrate in drinking water at different cage temperatures showed different interaction effects ($P < 0.05$) on the percentage of wing weight with a range of 14.69–19.14%. The results were consistent with (Irmayuanita *et al.*, 2015) where the percentage of back weight ranged from 15.33–17.41%. This was due to the influence of cage temperature and the concentration of fermented fruit filtrate on the increase in back weight percentage. Reduced feed intake led to a lower back weight percentage. Feed is the main factor influencing the percentage of carcass cuts in poultry, which include the breast, back, thigh, and wing (Merkley *et al.*, 1980). Nutrients such as protein, energy, and minerals are used to form bones, meat, and feathers based on the size and structure of wing feathers (Nurindah *et al.*, 2015).

CONCLUSION

In conclusion, acidifier of fermented star fruit filtrate and different cage temperatures after heat stress have a significant effect on production performance, carcass quality, commercial cut percentage, and blood hematology profile. Furthermore, the use of filtrate at a concentration of 1% with cage temperature of 32°C improves feed intake, body weight gain, live weight, carcass percentage, commercial cut percentage of the carcass, and blood hematology profile of KUB chickens.

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NOVELTY STATEMENT

This study specifically investigates fermented star fruit filtrate and its effects on local KUB chickens under factorial combinations of heat stress and dosage levels, which differs from previous studies conducted on broiler chickens. It not only targets a different genetic strain, but also integrates growth performance, carcass quality, and hematological responses during the post-stress recovery stage, offering a more comprehensive physiological insight that has not been previously reported.

AUTHOR'S CONTRIBUTION

FAA: Concept of research, laboratory work, data collection,

and data analysis.

AB and AMT: Laboratory work and data analysis.

NSA: Software, writing review and editing.

MA and DA: Edit and revise the manuscript.

GENERATIVE AI AND AI-ASSISTED TECHNOLOGY STATEMENT

Generative Artificial Intelligence (AI) and AI-Assisted Technology statement were not used in the preparation, analysis, or interpretation of the data presented in this manuscript. All parts of the paper, including the conceptual framework, research methodology, data analysis, discussion, and conclusion, were completely developed, written, and reviewed by the authors themselves.

CONFLICT OF INTEREST

The authors have declared no financial or institutional conflicts of interest related to the publication.

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