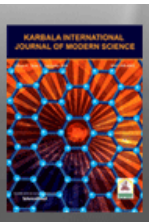

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Antibacterial Potential of Tapanuli Orangutan (Pongo tapanuliensis) Food in Batang Toru Forest Against Escherichia coli and Salmonella typhi

Herna Febrianty Sianipar
Department of Biology, Faculty of Mathematics and Natural Sciences, Brawijaya University, Jl. Veteran Malang 65133, East Java, Indonesia

Wahyu Widoretno
Department of Biology, Faculty of Mathematics and Natural Sciences, Brawijaya University, Jl. Veteran Malang 65133, East Java, Indonesia;

Luchman Hakim
Department of Biology, Faculty of Mathematics and Natural Sciences, Brawijaya University, Jl. Veteran Malang 65133, East Java, Indonesia;

Rezi Rahmi Amolia
Yayasan Ekosistem Lestari, Jl. Bunga Sedap Malam IX Medan 20132, North Sumatera, Indonesia.

Fatchiyah Fatchiyah
Research Center of Smart Molecule of Natural Genetics Resources, Brawijaya University, Jl. Veteran Malang 65133, East Java, Indo-nesia;, fatchiya@ub.ac.id

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Abstract

Diarrhea is a common disease affecting orangutans, primarily caused by *Escherichia coli* and *Salmonella typhi* bacteria. To treat this disease, antibacterial food sources are essential as therapeutic agents for orangutans. The fruits consumed by Tapanuli orangutans include *Campnosperma auriculatum, Agathis borneensis, Artocarpus heterophyllus, Castanopsis argantea,* and *Aglaia tomentosa.* This study aims to examine the amino acid and phytochemical components with potential antibacterial properties in these five fruit species and their inhibitory effects on *E. coli* and *S. typhi* growth through cell lysis, observed using a Scanning Electron Microscope (SEM). The samples were tested for amino acids, phytochemicals, vitamin C content, and antibacterial activity using the disc diffusion method, followed by observation of bacterial cells via SEM. *C. auriculatum* contained the highest amino acid levels, with significant values (pA. borneensis had high levels of L-Alanine, L-Threonine, L-Aspartic Acid, and L-Valine. Phytochemical analysis confirmed the presence of glycosides and vitamin C in all five samples, while leucoanthocyanidins were detected in *A. borneensis* and *A. tomentosa*. *C. auriculatum* had the highest vitamin C content (58.00 ± 8.76 mg/100g). The strongest antibacterial activity against *E. coli* was observed in *C. auriculatum* at a concentration of 125 mg/mL (18.7 mm), while *A. heterophyllus* exhibited the highest antibacterial activity against *S. typhi* at the same concentration (10.5 mm). SEM analysis confirmed that the extracts of *C. auriculatum* and *A. borneensis* inhibited the growth of *E. coli* and *S. typhi* bacteria through cell lysis. *C. auriculatum* demonstrated the highest potential as an antibacterial candidate.

Keywords

Amino Acid; Antibacterial; Fruits; Orangutans; Phytochemicals

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RESEARCH PAPER

Antibacterial Potential of Tapanuli Orangutan (*Pongo tapanuliensis*) Food in Batang Toru Forest Against *Escherichia coli* and *Salmonella typhi*

Herna F. Sianipar [a,b,c], Wahyu Widoretno [a], Luchman Hakim [a], Rezi R. Amolia [d], Fatchiyah Fatchiyah [a,b,*]

[a] Department of Biology, Faculty of Mathematics and Natural Sciences, Brawijaya University, Jl. Veteran, Malang 65133, East Java, Indonesia
[b] Research Center of Smart Molecule of Natural Genetics Resources, Brawijaya University, Jl. Veteran, Malang 65133, East Java, Indonesia
[c] Department of Water Resource Management, Faculty of Engineering and Water Resource Management, Universitas HKBP Nommensen Pematangsiantar, Jl. Siopat Suhu, Pematangsiantar 21136, North Sumatera, Indonesia
[d] Yayasan Ekosistem Lestari, Jl. Bunga Sedap Malam IX, Medan 20132, North Sumatera, Indonesia

Abstract

Diarrhea is a common disease affecting orangutans, primarily caused by *Escherichia coli* and *Salmonella typhi* bacteria. To treat this disease, antibacterial food sources are essential as therapeutic agents for orangutans. The fruits consumed by Tapanuli orangutans include *Campnosperma auriculatum, Agathis borneensis, Artocarpus heterophyllus, Castanopsis argantea,* and *Aglaia tomentosa*. This study aims to examine the amino acid and phytochemical components with potential antibacterial properties in these five fruit species and their inhibitory effects on *E. coli* and *S. typhi* growth through cell lysis, observed using a Scanning Electron Microscope (SEM). The samples were tested for amino acids, phytochemicals, vitamin C content, and antibacterial activity using the disc diffusion method, followed by observation of bacterial cells via SEM. *C. auriculatum* contained the highest amino acid levels, with significant values ($p < 0.05$) for L-Glutamic acid, L-Phenylalanine, L-Arginine, and L-Tyrosine. Meanwhile, *A. borneensis* had high levels of L-Alanine, L-Threonine, L-Aspartic Acid, and L-Valine. Phytochemical analysis confirmed the presence of glycosides and vitamin C in all five samples, while leucoanthocyanidins were detected in *A. borneensis* and *A. tomentosa*. *C. auriculatum* had the highest vitamin C content (58.00 ± 8.76 mg/100 g). The strongest antibacterial activity against *E. coli* was observed in *C. auriculatum* at a concentration of 125 mg/mL (18.7 mm), while *A. heterophyllus* exhibited the highest antibacterial activity against *S. typhi* at the same concentration (10.5 mm). SEM analysis confirmed that the extracts of *C. auriculatum* and *A. borneensis* inhibited the growth of *E. coli* and *S. typhi* bacteria through cell lysis. *C. auriculatum* demonstrated the highest potential as an antibacterial candidate.

Keywords: Amino acid, Antibacterial, Fruits, Orangutans, Phytochemicals

1. Introduction

Lack of or poor-quality food sources causes concerning conditions for many animals, including orangutans. Such conditions lead to stress, which impacts the immune response and increases susceptibility to infection. These infections are primarily caused by gastrointestinal pathogens. One major cause of disease in orangutans is pathogenic bacteria [1,2]. Among these, *Escherichia coli* and *Salmonella spp.* are commonly identified pathogens, having been isolated from primates feces [3].

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* Corresponding author at: Department of Biology, Faculty of Mathematics and Natural Sciences, Brawijaya University, Jl. Veteran, Malang, 65133, East Java, Indonesia.
E-mail addresses: hernasianipar21@gmail.com (H.F. Sianipar), wahyu_widoretno@yahoo.com (W. Widoretno), luchman@ub.ac.id (L. Hakim), rezi.rahmi@yel.or.id (R.R. Amolia), fatchiya@ub.ac.id (F. Fatchiyah).

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After amoebic dysentery, salmonellosis is the second most serious disease affecting orangutans in Bukit Lawang. Infections can occur through contact with humans, contaminated water, or tourist waste. *Salmonella typhi* is a major cause of diarrhea in orangutans, with seven out of 28 clinical cases resulting in death [4,5]. Since orangutans rely solely on their diet, their food is believed to contain bioactive compounds with potential therapeutic effects against infectious diseases [6]. Research identified *Ficus benjamina* and *Ficus elastica* as the most effective among 34 primate food plants in exhibiting antibacterial activity against *Bacillus subtilis* [7]. Conservation efforts utilizing local resources, such as orangutan food, have been explored in previous studies [8]. In the Batang Toru Forest, five species of fruit have been recorded as preferred and primary food sources of Tapanuli orangutans, with the fruiting season occurring from October to December: *Campnosperma auriculatum, Agathis borneensis*, *Artocarpus heterophyllus*, *Castanopsis argantea,* and *Aglaia tomentosa* [9]. The highest carbohydrates are found in *C. argentea* (84.09 %), protein in *A. tomentosa* (8.52 %), fat in *C. auriculatum* (3.73 %), energy from fat *in C. auriculatum* (33.57 Kcal/100 g), total energy in *C. argantea* (386.34 Kcal/100 g), ash content in *A. borneensis* (5.32 %) and water content in *A. borneensis* (15.98 %). The monkey diet mostly consists of plant-based foods, especially fruits (33.5 %), while orangutans consume fruit (71.06 %). Consumption of animal protein is 54.8 % in monkeys and 16.3 % in orangutans [10–12].

A. heterophyllus fruit is generally consumed and has been used to treat tuberculosis, spleen disorders, dysentery, malaria, and diarrhea [13]. Various components of the jackfruit tree are utilized in traditional medicine. Jackfruit contains polyphenols and tannins, both of which exhibit antibacterial activity [14]. Research showed that ethanol extract of jackfruit demonstrates antibacterial activity against *E. coli* and *Staphylococcus aureus*, producing an inhibitory zone diameter classified as moderate [15]. An antibacterial test of Bornean orangutan feed derived from *Eusideroxylon zwageri* bark showed inhibition of the growth of *Cutibacterium acnes* (16.3 mm), *S. typhi* (13.8 mm), *S. aureus* (13.9 mm), and *Candida albicans* (13.9 mm) [16]. Other bioactive compounds, such as vitamin C, are essential in strengthening the body's resistance to infections. Orangutans cannot synthesize vitamin C due to the absence of the enzyme required to convert L-gluconic acid into ascorbic acid; therefore, they must obtain it from their diet. Vitamin C functions as an antioxidant, a key component of collagen synthesis, and an antibacterial agent [17]. It is known to inhibit the enzyme activity of *Streptococcus pneumoniae,* thereby reducing the spread of this pathogen during the initial phase of pneumococcal invasion. In addition, vitamin C also contributes to lowering intestinal pH, which inhibits the growth of pathogenic bacteria that are intolerant to acidic conditions [18–20]. Amino acids, such as lysine, also possess antibacterial properties. Lysine inhibits bacterial growth by binding to and inactivating enzymes involved in the synthesis of bacterial cell wall. This weakens the bacterial cell walls, making them more susceptible to damage [21]. Given these concerns, research on orangutan dietary components is necessary to prevent the proliferation of harmful bacteria such as *E. coli* and *S. typhi*. This research aims to analyze the phytochemical compounds and amino acid composition as well as evaluate the antibacterial activity of Tapanuli orangutan (*Pongo tapanuliensis*) food sources from the Batang Toru forest against *E. coli* and *S.typhi*.

2. Methods

2.1. Plant material

This research was conducted from October 2023 to May 2024 at the Sustainable Ecosystem Foundation Orangutan Conservation Program Research Station in the Batang Toru Forest Area (Camp Mayang), North Sumatra Province. The fruit species *C. auriculatum*, *A. borneensis*, *A. heterophyllus*, *C. argantea*, and *A. tomentosa* were selected according to their fruiting seasons from October to December. Laboratory tests were conducted in several facilities, including PT. Saraswanti Indo Genetech and the Bioscience Laboratory Center, Brawijaya University.

2.2. Preparation and extraction

Two hundred grams of the ripe fruits from *C. auriculatum, A. borneensis*, *A. heterophyllus*, *C. argantea,* and *A. tomentosa* were washed with clean water and dried in an oven at 60 °C for 36 h. The dried fruits were then ground using a blender into simplicia powder. Ten grams of simplicia powder was weighed, mixed with 100 ml of 96 % ethanol, and filtered after extraction. The extract was subsequently concentrated using a rotary evaporator set at 40 °C and 90 rpm [22].

2.3. Determination of amino acids

Amino acid levels were determined using fruit powder. UPLC was employed to analyze L-Alanine, L-Aspartic Acid, L-Phenylalanine, L-Arginine, L-Threonine, L-Glutamic Acid, L-Glycine, L-Valine, L-

Proline, L-Serine, and L-Tyrosine following Protocol 18-5- 17/MU/SMM-SIG. The analysis was performed using an AccQ, tag ultra C18 1.7 μm column (2.1 × 100 mm) was used for UPLC. The mobile phase consisted of Eluent A (Eluent A concentrate), Eluent B (AccQ.Tag Ultra Amino Acid Analysis 10 % in water), Eluent C (Aquabides), and Eluent D (AccQ.Tag Ultra Amino Acid Analysis). The flow rate was established at 0.5 mL/min, and detection was performed using a photodiode array (PDA) detector at 260 nm. (An AccQ.Tag Ultra Columns and Xevo TQ-S Micro triple quadrupole mass spectrometer Waters, USA and merck (Sigma-Aldrich) amino acid standard were used in this study) [22].

2.4. Phytochemicals test

Extracts of *C. auriculatum*, *A. borneensis*, *A. heterophyllus*, *C. argantea*, and *A. tomentosa* were subjected to phytochemical tests using Whatman No. 1 filter paper. The filtrate was analyzed to determine phytochemical content, with positive results indicated by a color change: glycosides (yellow or red), leucoanthocyanidins (brown or red), and vitamin C (clear or slightly yellow) [22].

2.5. Vitamin C

One mL of the sample was mixed with 9 mL of d 2.6 - dichlorophenol indopenol, homogenized, and its absorbance measured at 516 nm. The reagents used in this study included iodine, and ascorbic acid was used as the standard solution [23].

2.6. Antibacterial activity

Pure cultures of *E. coli* and *S. typhi* were obtained from the Microbiology Laboratory of Brawijaya University. The bacteria were cultivated in Luria-Bertani (LB) broth (Bioworld) at 37 °C overnight, reaching a density of 10^8 cells/mL. A UV−vis spectrophotometer (SmartSpec PlusTM, Bio-Rad Laboratories Inc., Hercules, CA, USA) was utilized to measure absorbance at 600 nm. The disc diffusion technique was employed to evaluate antibacterial activity. Bacterial cultures were transferred to Mueller-Hinton medium, and 6 mm paper discs were soaked with 0.1 mL of the sample extract. The zone of inhibition was measured after 1080 min of incubation at 37 °C. SEM analysis was performed to examine the effect of the extract on bacterial cells, with modifications based on previous studies. A total of 150 μL of extract and 150 μL of bacterial culture were added to 15 mL of LB medium and incubated for 18 h at 37 °C. The cultures were then centrifuged at 2000×*g* for 10 min at 4 °C. After two washes with 0.9 % sodium chloride, the pellet was fixed in 3 % glutaraldehyde for 30 min. Samples were washed three times for 15 min each with phosphate-buffered saline and dehydrated using a graded ethanol series (30, 50, 70, 80, 90, and 96 %). Platinum-coated slides were then examined using SEM (Hitachi TM3000; Tokyo, Japan) [24].

2.7. Data analysis

Statistical analysis was performed to compare the content of amino acids, phytochemical test, vitamin C, and antibacterial activity in Tapanuli orangutan foods (*C. auriculatum*, *A. borneensis*, *A. heterophyllus*, *C. argantea*, and *A. tomentosa*). A t-test was applied at a 95 % confidence level. To assess variations among the five foods and identify significant differences, a one-way ANOVA was conducted followed by Tukey's post hoc test at a 95 % confidence level ($p < 0.05$). Data were analyzed using SPSS version 16.0 and GraphPad Prism 8 [25].

3. Result and discussion

3.1. Amino acid content

Amino acids are important biomolecules that serve as the fundamental building blocks of proteins [24]. As shown in Table 1, the amino acid composition varies significantly among the five plant species consumed by the Tapanuli orangutan. *C. auriculatum* exhibits the highest levels of L-Arginine (4760.907 ppm), L-Glycine (3950.61 ppm), L-Phenylalanine (2432.055 ppm), L-Glutamic Acid (7508.4 ppm) and L-Tyrosine (1854.22 ppm). Meanwhile, *A. borneensis* contains the highest concentrations of L-Alanine (3612.293 ppm), L-Aspartic Acid (8716,797 ppm), L-Valine (3103.78 ppm), L-Proline (2892.327 ppm), and L-Threonine (2310.938 ppm). *C. argantea, A. heterophyllus,* and *A. tomentosa* have comparatively lower amino acid contents.

Arginine is a positively charged amino acid that can bind to negatively charged bacterial cell surface components such as lipopolysaccharide (LPS), teichoic acid, and phospholipids, demonstrating antibacterial potential. Arginine solutions at concentrations of 5 %, 8 %, 10 %, and 12 % exhibit antibacterial power against *Enterococcus faecalis*, with higher concentrations exhibiting greater efficacy [26,27].

3.2. Phytochemical profile

A phytochemical test was conducted to identify bioactive compounds in the ethanol extracts of

*Table 1. Amino Acid Content in food of Tapanuli orangutan (*Pongo tapanuliensis*).*

Component Amino acid	Concentration (ppm)				
	C. auriculatum	*A. borneensis*	*C. argantea*	*A. heterophyllus*	A. tomentosa
L-Alanine	3556.57[b]	3612.29[a]	3177.25[c]	2047.95[e]	2804.63[d]
L-Arginine	4760.90[a]	3566.00[b]	3448.15[c]	2820.17[d]	2302.47[e]
L-Aspartic Acid	7873.09[b]	8716.79[a]	6077.28[d]	6112.16[c]	5358.71[e]
L-Glycine	3950.61[a]	3700.52[b]	2451.09[c]	1596.07[e]	2310.15[d]
L-Glutamic Acid	7508.40[a]	5694.95[b]	5510.85[d]	2899.45[e]	5571.04[c]
L-Valine	2370.16[b]	3103.78[a]	1877.23[c]	1606.63[e]	1838.69[d]
L-Phenylalanine	2432.05[a]	2110.99[b]	2052.30[d]	1360.90[e]	2067.95[c]
L-Proline	2854.06[b]	2892.32[a]	1917.87[d]	1452.20[e]	2412.25[c]
L-Serine	2596.36[b]	2354.84[c]	2126.93[d]	1486.52[e]	2759.24[a]
L-Threonine	1819.38[b]	2310.93[a]	1694.08[d]	1126.68[e]	1746.24[c]
L-Tyrosine	1854.22[a]	1643.57[b]	1245.32[d]	857.62[e]	1549.45[c]

Note: Different letters indicate significant differences of effects amongs fruit species (Tukey's HSD; $P < 0.05$).

Tapanuli orangutan food samples. Positive results were indicated by a color change, while negative results showed no change. As shown in Table 2, all five samples tested positive for glycosides and leucoanthocyanidins, with *C. auriculatum* showing the highest qualitative test results. The other four samples, *A. borneensis*, *A. heterophyllus*, *C. argantea*, and *A. tomentosa*, showed similar results.

Aglaia elliptica, *Croton argyratus*, *Artocarpus lanceifolius*, *Artocarpus odoratissimus*, and *Baccaurea macrocarpa,* which are food sources of Bornean orangutans, tested positive for alkaloids, flavonoids, tannins and steroids. While the food of Tapanuli orangutans, such as *C. auriculatum*, *A. borneensis*, *C. argentea*, *A. heterophyllus*, *A. tomentosa,* tested positive for alkaloids, flavonoids, tannins, phenolics, and quinolines [10,11,28].

Glycosides are compounds composed of sugars and aglycones (non-sugar parts) that exhibit various biological activities, including antibacterial properties. Certain glycosides, such as flavonoid glycosides and saponins, have been shown to inhibit the growth of pathogenic microorganisms. Glycosides belong to the alkaloid compound group and share a similar antibacterial mechanism of action with alkaloids [29,30]. Previous LC-MS/MS analysis reported that glycosides made up 99.82 % of the compounds in *A. tomentosa* fruit [11]. The inhibitory mechanism involves disrupting peptidoglycan synthesis in bacterial cells, leading to incomplete cell

*Table 2. Phytochemicals in food of Tapanuli orangutan (*Pongo tapanuliensis*).*

Sample	Glycosides	Observation Glycosides	Leucoanthocyanidins	Observation Leucoanthocyanidin
C. auriculatum	+++		++	
A. borneensis	++		+	
C. argantea	++		+	
A. heterophyllus	++		+	
A. tomentosa	++		+	

Note: (−) absence of targeted substance, (+) low intensity of targeted substance, (++) moderate intensity of targeted substance, (+++) high intensity of targeted substance.

wall formation and eventual cell death. Leucoanthocyanidins, consisting of two flavonoid units bound to a sugar group, influence both physical and biochemical properties [31]. Their antibacterial activity functions by damaging bacterial cell membranes, causing leakage and subsequent cell death [32].

3.3. Vitamin C content

Table 3 shows that *C. auriculatum* fruit has the highest vitamin C content (58 mg/100 g), while *A. heterophyllus* has the lowest (29.06 mg/100 g). Statistical analysis reveals no significant differences in vitamin C content among *C. auriculatum*, *A. borneensis*, and *C. argantea* indicating similar vitamin C content in these three fruits.

Vitamin C acts as an antioxidant by producing Reactive Oxygen Species (ROS), which are highly reactive oxygen-derived oxidizing compounds consisting of radical and non-radical groups [33]. In *Streptococcus pneumoniae,* vitamin C binds to Fe, generating hydroxyl radicals through a combination of the Haber–Weiss cycle and the Fenton reaction [34]. ROS attack key bacterial structure by damaging the lipid layer of the cell membrane, disrupting metabolism and replication. Osmotic pressure causes water influx into bacterial cells, leading to lysis and cell death. Previous research showed that the binding of vitamin C to iron in *Mycobacterium tuberculosis* generates hydroxyl radicals, leading to bacterial DNA damage [35,36].

3.4. Antibacterial activity

Table 4 shows that *C. auriculatum* at a concentration of 125 mg/mL exhibits the highest inhibition zone against *E. coli* (18.7 mm), classified as strong inhibition. No inhibition zone was observed for *S. typhi*, likely due to differences in bacterial susceptibility to antibacterial compounds. *S. typhi* has a stronger defense mechanism, including a thicker outer membrane compared to some *E. coli* bacteria, and may cause antagonistic interactions with bioactive compounds, reducing antibacterial efficacy. As a result, *C. auriculatum* is less effective against *S. typhi* [37]. For *S. typhi*, *A. heterophyllus* at 125 mg/mL demonstrates the highest inhibition zone (10.5 mm). Catechin, a flavonoid found in higher concentrations in *A. heterophyllus* extract, reported to inhibit the growth of *Pseudomonas aeruginosa*, *E. coli*, *Streptococcus mutans*, and *Salmonella typhimurium* [38]. Statistical analysis indicates significant differences among the five samples for *E. coli* at 125 mg/mL and 250 mg/mL. However, for *S. typhi* at 250 mg/mL, *A. borneensis* and *A. heterophyllus* show similar inhibition zones (Table 4).

According to Ref. [39], antibacterial strength is classified based on the diameter of the inhibition

*Table 3. Vitamin C content in food of Tapanuli orangutan (*Pongo tapanuliensis*).*

Sample	Qualitative screening	Observation Vitamin C	Vitamin C (mg/100 g)
C. auriculatum	++		58.00 ± 8.76[a]
A. borneensis	+		49.36 ± 3.44[a]
C. argantea	+		42.4 ± 4.44[a]
A. heterophyllus	+		1.55 ± 1[c]
A. tomentosa	+		29.06 ± 3.7[b]

Note: Different letters indicate significant differences of effects amongs fruit species (Tukey's HSD; $P < 0.05$).

Table 4. Antibacterial activity results in test of Tapanuli orangutan food against E. coli *and* S. typhi.

Sample	Inhibition zone diameter (mm)					
	E. coli			*S. typhi*		
	0 mg/mL	125 mg/mL	250 mg/mL	0 mg/mL	125 mg/mL	250 mg/mL
C. auriculatum	0^{c}	18.7^{Aa}	17.5^{Ab}	0^{c}	0^{Ea}	0^{Da}
A. borneensis	0^{c}	13.9^{Da}	12^{Cb}	0^{c}	9.7^{Ca}	7.9^{Bb}
C. argantea	0^{c}	15^{Ca}	10.5^{Db}	0^{c}	10.2^{Ba}	9^{Ab}
A. heterophyllus	0^{c}	11.5^{Ea}	8.3^{Eb}	0^{c}	10.5^{Aa}	7.9^{Bb}
A. tomentosa	0^{c}	16.5^{Ba}	13.9^{Bb}	0^{c}	7^{Da}	6.2^{Cb}

Note: Different letters indicate significant differences of effects amongs fruit species (Tukey's HSD; $P < 0.05$).

zone as follows: ≤5 mm (weak), 5–10 mm (moderate), 10–20 mm (strong), and ≥20 mm (very strong). Based on this classification, Robusta coffee extract displayed very strong antibacterial activity against *E. coli* at concentrations of 10 % (22.5 mm), 50 % (24 mm), and 100 % (27 mm), indicating their effectiveness in inhibiting *E. coli* growth.

The effect of *C. auriculatum* and *A. borneensis* extracts at concentrations of 125 and 250 mg/mL on bacterial cells was observed using SEM. The morphology of *E. coli* and *S. typhi* bacteria is presented in Figs. 1 and 2. Untreated *E. coli* cells (0 mg/mL) measured 2.16–2.58 μm in *C. auriculatum* and 1.6–2.45 μm in the *A. borneensis* group. After treatment with *C. auriculatum* (125–250 mg/mL), *E. coli* cell lengths decreased to 901 nm–2.05 μm, while treatment with *A. borneensis* resulted in lengths of 972 nm–2.14 μm, indicating a cell size reduction following exposure to the extracts.

The cell length of untreated *S. typhi* (0 mg/mL) ranged from 1.71 to 2.91 μm in the *C. auriculatum* and 1.18–3.41 μm in the *A. borneensis* group. After treatment with *C. auriculatum* extract (125–250 mg/mL), *S. typhi* cell lengths decreased to 924 nm–1.48 μm, while treatment with *A. borneensis* resulted in lengths of 1.23–1.51 μm, indicating a reduction in bacterial cell size after exposure to both extracts.

Morphological changes in bacterial cells occur due to lysis, caused by the loss of cell wall integrity in the presence of bioactive compounds [40]. Fusion of *S. typhi* bacterial cells treated with 125 mg/mL A. *borneensis* extract was observed, with visible melting indicating severe damage leading to cell death. Based on the inhibition zone and bacterial cell length analysis, Tapanuli orangutan food extracts were more efficient in suppressing *E. coli* proliferation compared to *S. typhi* [41].

The greater effectiveness of Tapanuli orangutan food extract against *E. coli,* a Gram-negative bacterium can be attributed to its relatively thin peptidoglycan layer (approximately 2–3 nm) [42], which is surrounded by an outer membrane containing LPS [43]. This structural characteristic makes *E. coli* more susceptible to certain bioactive compounds. In contrast, *S. typhi,* also a Gram-negative bacterium, has a more complex and thicker cell wall than *E. coli,* with more variations in its LPS layer, which may contribute to increased resistance to some antimicrobial compounds [44].

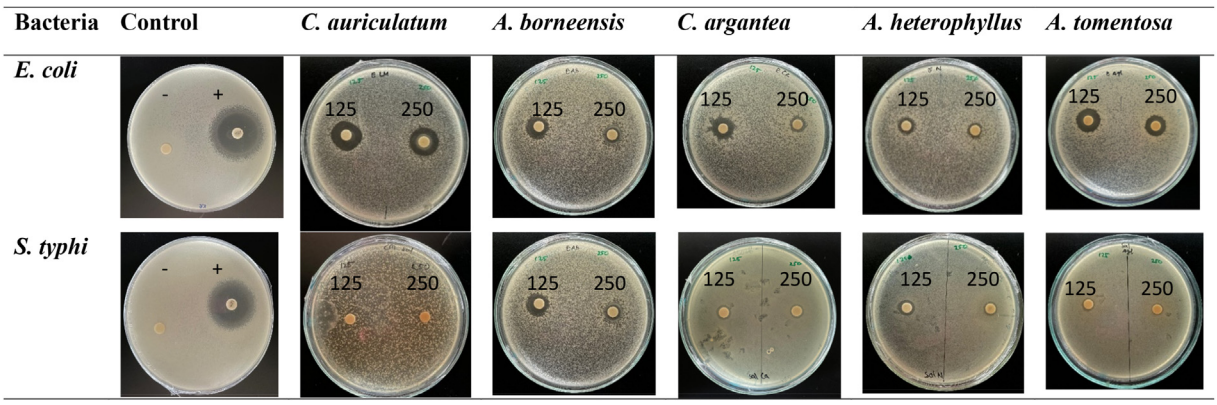

Fig. 1. Antibacterial activity results in test of Tapanuli orangutan food against E. coli *and* S. typhi *Concentrations 125 and 250 mg/mL with positive control (chloramphenicol) and negative control (aquadest).*

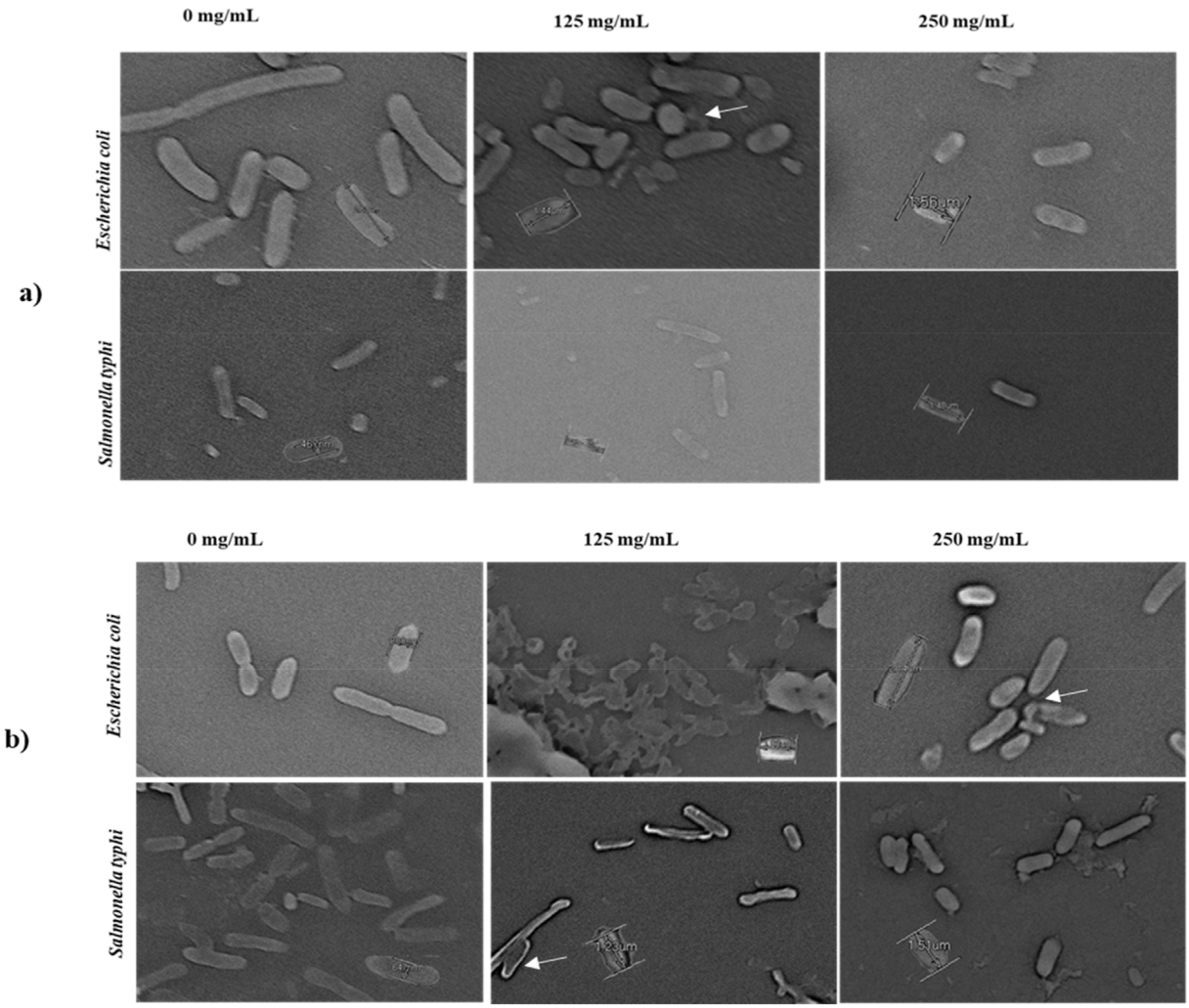

Fig. 2. Morphology bacterial scanning electron microscope E. coli *and* S. typhi *(12000x magnification) as effect by a)* C. auriculatum *extract; b)* A. borneensis *extract. The white arrow indicates lysis.*

4. Conclusion

The five Tapanuli orangutan food samples—*C. auriculatum*, *A. borneensis*, *A. heterophyllus*, *C. argantea*, and *A. tomentosa*—exhibited antimicrobial activity against *E. coli* and *S. typhi*. Among them, *C. auriculatum* showed the highest antibacterial potential. Developing conservation strategies for Tapanuli orangutan food should account for the ecological impact of harvesting these fruits for therapeutic use, as well as conducting in vivo studies using animal models infected with diarrhea. These efforts would support in vitro and pharmacological tests, which are essential for the continued advancement of this research.

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Ethics information

All plant materials used in this study were identified and authenticated by the UPT Herbal Materia Medica Laboratory, Batu, East Java Province, Indonesia. The identification reference numbers are as follows: *C. auriculatum* (No. 0009.3/137/102.20/ 2024), *A. borneensis* (No. 0009.3/134/102.20/2024), *C. argantea* (No. 0009.3/138/102.20/2024), *A. heterophyllus* (No. 0009.3/136/102.20/2024), and *A. tomentosa* (No. 0009.3/135/102.20/2024).

Conflicts of interest

The authors declared that there are no conflicts of interest.

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