The effectiveness of coconut water in inhibiting *Shigella* sp. bacteria from diarrhea

Ikhsan Mujahid ¹, Arif Mulyanto, Tyas Ulfah Khasanah ²

¹ Medical Laboratory Technology, Universitas Muhammadiyah Purwokerto, Purwokerto, Jawa Tengan, Indonesia
² The Faculty of Education, Universitas Muhammadiyah Purwokerto, Purwokerto, Jawa Tengan, Indonesia

**ABSTRACT**

**Background:** Diarrhea is still a serious problem for some people. It can be affected by the *Shigella* sp. bacteria. Some villagers drink tender coconut water (*Co-cos nucifera L*) to cure diarrhea. Some types of consumed tender or green coconut are green coconut, brown coconut, *wulung* green coconut, and *wulung* brown coconut. It is necessary to determine the contribution of coconut types to treat diarrhea caused by the *Shigella* sp bacterium.

**Objective:** to determine the benefit of several types of coconut water toward diarrhea caused by bacteria such as *Shigella* sp.

**Method:** The method implemented was to determine the benefit of tender coconut water toward *Shigella* sp bacteria was experimental study through Kirby-Bauer test (antibiotic disc). The study selected water of green coconut, brown coconut, *wulung* green coconut, and *wulung* brown coconut, while the positive control used *Lodia* diarrhea drug and negative control used aquadest (Purified water). Testing data of green coconut water toward *Shigella* sp. was analyzed through analysis of variance (ANOVA) with F test at 95% significant level. Then it was continued by Duncan test at 95% significant level.

**Results:** The results of testing some green coconut water, positive control and negative control on *Shigella* sp bacteria can be seen from the clear zone formed around the antibiotic disc, green coconut showed 6.63 mm, brown coconut was 6.57 mm, *wulung* green coconut was 16.63 mm, *wulung* brown coconut was 9.23 mm, *Lodia* diarrhea drug was 12.33 mm and purified water was 7.27 mm.

**Conclusion:** all green coconut water is able to inhibit the growth of *Shigella* sp. The best inhibitor is obtained from the administration of *wulung* green coconut water with 16.6 mm diameter of the inhibition zone.

**KEYWORDS**

Diarrhea; Anti-Bacterial Agents; *Shigella* sp bacteria; Coconut

**INTRODUCTION**

Diarrhea is still a serious problem for some people. Diarrhea can be affected from several organisms such as the Rotavirus, Adenovirus and some bacteria such as *Shigella* sp, *Escherichia coli*, *Vibrio cholerae*, and other bacteria. Besides, diarrhea can be stimulated by malabsorption, parasites, allergies, immunodeficiency and food poisoning ¹. Extraordinary event (KLB) of diarrhea occurred in 2015, there were 18 incidents which occurred in 11 provinces, 18 districts/cities with 1,213 number of sufferers and caused 30 people die (2.47%) ².

Diarrhea caused by *Shigella* sp in Indonesia was relatively high which caused 29% of dysentery sufferers die ³. If no treatment given to dysentery sufferers, there will be high possibility to be serious and even death ⁴.

Some villagers consume tender coconut water to treat diarrhea. It is now becoming popular to have medicinal tender coconut (*wulung* green coconut) to heal some illnesses such as diarrhea. It shows that some are recovered and some are reducing the intensity from diarrhea after drinking the tender coconut water ⁵. Based on the study ⁶, tender coconut water is commonly used to treat diarrhea suf-
ferers. Besides, this water is also beneficial to cure intestinal worm and cholera, to reduce itching due to small pox and some other skin diseases.

Currently, it is familiar with medicinal coconut or people called as wulung coconut. This coconut has a pink color in the mesocarp (coir). On the outer skin (epicarp) the color is not always green but sometimes brownish. Wulung coconut is able to cure various types of diseases. Therefore, people have been using coconut water for a long time to treat poisoning. Even wulung coconut water is also believed to be able to cure Hand, Foot and Mouth disease.

Based on the available strengths, it is necessary to conduct a test to tender coconut water in curing diarrhea caused by Shigella sp. bacteria.

METHOD

Research materials
The instruments used in this study were autoclave, Laminar Air Flow (LAF), micropipette, petri dish, and test tube. The materials used included alcohol, Nutrient Agar (NA) media, Nutrient Broth (NB) media, antibiotic disc, green coconut, brown coconut, wulung green coconut, wulung brown coconut, lodia diarrhea drug as positive control and purified water as negative control.

Experimental design
This study was an experiment to reveal the benefit of tender coconut water to Shigella sp bacteria by Kirby Bauer method (antibiotic disc). The diameter of the inhibition zone formed was measured at the time of observation.

Working procedure and research variables

Bacterial bejuvenation
Bacterial rejuvenation was carried out by inoculating 1 needle from Shigella sp bacteria on NB medium and incubating for 1x24 hours at 37 °C.

Treatment preparation
The extraction from coconut water from each green coconut, brown coconut, wulung green coconut and wulung brown coconut was placed into a sterile test tube and treated aseptically. Positive control was done by adding 1 gram of lodia with 10 ml of sterile purified water, while the negative control used sterile purified water. A sterile NA medium and other equipments were sterilized.

Kirby Bauer test (antibiotic disc)
Sterile NA medium was poured into sterile petri dishes. 100 µl rejuvenated Shigella sp bacteria was placed on a spread plate and flattened with drugalsky. Then, each 3 blank discs were inserted with dripped coconut water (green coconut, brown coconut, wulung green coconut, wulung brown coconut), for positive control was dripped with Lodia diarrhea drug and negative control was sterile purified water. Next, the inhibition zone was observed and measured formed around the antibiotic disc.

Research analysis
The results of inhibitory zone diameters measurements were analyzed through analysis of variance (ANOVA). If there was a significant difference, Duncan Multiple Range (DMRT) test was continued at the 95% significant level and explained descriptively.

RESULTS
The results of the average diameter in inhibition zone of each treatment were as follow; green coconut at 6.63 ± 0.25 mm, brown coconut at 6.57 ± 0.51 mm, wulung green coconut at 16.63 ± 4.84 mm, and wulung brown coconut at 9.23 ± 1.96 mm. The average diameter of inhibition zone for positive controls (lodia diarrhea drug) was 12.33 ± 1.01 mm and negative control (purified water) was 7.27 ± 1.32 mm (Table 1). The diameter of the inhibitory zone for each treatment can be seen in Figure 1.

The results indicated that the treatment of green coconut, brown coconut, wulung green coconut, wulung brown coconut, and purified water presented no significant difference in affecting the growth of Shigella sp. In the treatment of wulung brown coconut and diarrhea drug (lodia) illustrated no significantly different, and the diameter of the inhibition zone was wider than that of green coconut water. Whereas, the treatment of wulung green coconut water was significantly different from the other treatments. It could be seen from the extent of the inhibition zone created.

DISCUSSION
The treatment of wulung green coconut contributed the widest inhibiting zone compared to other treatments. The ability of coconut water to inhibit the growth of Shigella sp bacteria was due to the presence of tannin compounds found in coconut water. It was supported by 5, that tannin and antidote (anti-toxin) in wulung coconut water could inhibit bacterial growth.

The mechanism of tannin in inhibiting Shigella sp. was by terminating the cell membrane, inactivating essential enzymes, and decoding the function of genetic material. Meanwhile, according to 10, the mechanism of tannin as an antibacterial was to inhibit the reverse transcriptase enzymes and DNA topoisomerase so that bacterial cells would not appear. Besides, tannins have anti-bacterial activities related to their ability to activate adhesion of microbial cells as well as enzymes, and interfere protein transport in the inner cell layer. The presence of tannins in green coconut water contributed to inhibit the growth of Shigella. Sp. as one of the bacteria which causes diarrhea.
Table 2. The result of inhibiting zone diameter in some coconut water, diarrhea medicine and purified water toward shigella sp. bacteria.

| Intervention                  | Inhibiting zone diameter of Shigella sp. bacteria (mm) | Average diameter of inhibiting zone (mm) |
|-------------------------------|-------------------------------------------------------|----------------------------------------|
|                               | T₁          | T₂          | T₃          |                                            |
| Wulung green coconut          | 21.8        | 15.9        | 12.2        | 16.63±4.84<sup>a</sup>                     |
| Wulung brown coconut          | 11.0        | 9.6         | 7.1         | 9.23±1.98<sup>bc</sup>                     |
| Green coconut                 | 6.9         | 6.4         | 6.6         | 6.63±0.25<sup>c</sup>                      |
| Brown coconut                 | 7.0         | 6.0         | 6.7         | 6.57±0.51<sup>c</sup>                      |
| Lodia diarrhea drug           | 11.4        | 13.4        | 12.2        | 12.33±1.01<sup>b</sup>                     |
| Aquades                       | 7.0         | 6.1         | 8.7         | 7.27±1.32<sup>c</sup>                      |

T= test, a = strong inhibiting, b = moderate inhibiting, c = weak inhibiting

For negative control, sterile purified water, the test results obtained clear zone around the antibiotic disc. The inhibition was due to the occurrence of water osmosis into bacterial cells which caused the bacterial cells to experience lysis. It was supported by 12, which stated that one of the causes of lysis cells was the imbalance of osmotic pressure between environmental and cell pressures. If the environmental conditions were more hypotonic than the pressure conditions in the cell, or the conditions in the cell are more hypertonic than the environmental conditions, the cell would experience lysis. As a result, the cells would develop and eventually break.

As a positive control, the Lodia diarrhea drug was selected which was able to form inhibitory zone against Shigella sp. The active compound of Lodia was Loperamide HCl. Lodia was an antibiotic for diarrhea which worked by inhibiting bacterial cell wall synthesis. Lodia worked directly on cell membranes of microorganisms, increased permeability and caused leakage of intra-cellular compounds, interfered with the function of 30 S or 50 S ribosomal units to inhibit reversible protein synthesis, binded to 30 S ribosome subunits and interfered with protein synthesis, affected bacterial nucleic acid metabolism, and retained important enzymes from folate metabolism.

According to 13, stated that Loperamide was antimotility drug. Meanwhile 14 the way Loperamide worked was to slow down intestinal motion and make stools more dense. Loperamide was also used to reduce the amount of fluid which came out in patients who had an ileostomy, and to treat diarrhea in people with inflammatory bowel disease.

CONCLUSIONS AND RECOMMENDATION

Coconut water is able to inhibit Shigella sp bacteria which causes diarrhea. Green coconut water, brown coconut water, wulung brown coconut water, wulung green coconut water, diarrhea (lodia) and purified water can inhibit the growth of Shigella sp. Meanwhile, the wulung green coconut water was the most effective and significant to inhibit the growth of the bacteria compared to other treatments with the diameter of the inhibition zone is 16.6 mm. Further research is required on what levels of tannins and types of specific compounds exist in green coconut water.

REFERENCES

1. Widoyono MPH. Penyakit Tropis Epidemiologi, Penularan, Pencegahan & Pemberantasan. Jakarta; 2011.
2. Kementerian K. Data dan Informasi Tahun 2014 (Profil Kesehatan Indonesia). Kementerian Kesehatan Republik Indonesia. 2015.

3. Nafianti S, Sinuhaji AB. Resisten Trimetoprim – Sulfametoksazol terhadap Shigellosis. Sari Pediatr. 2017;7(1):39. doi:10.14238/sp7.1.2005.39-44

4. Yusuf S. Profil Diare di Ruang Rawat Inap Anak. Sari Pediatr. 2011;13(4):265-270. https://saripediatri.org/index.php/sari-pediatri/article/download/424/356.

5. putri soraya mansur. khasiat kelapa hijau bertampuk merat atau wulung. khasiat kelapa hijau bertampuk merat atau wulung. http://puan.co/2017/04/khasiat-kelapa-hijau-bertampuk-merah-atau-kelapa-wulung/ Published 2017.

6. Chan E, Elevitch CR. Cocos nucifera (coconut). Species profiles Pacific Isl Agrofor. 2006;2:1-27.

7. Barlina R. Potensi buah kelapa muda untuk kesehatan dan pengolahannya. Perspektif. 2016;3(2):46-60.

8. Yong JW, Ge L, Ng YF, Tan SN. The chemical composition and biological properties of coconut (Cocos nucifera L.) water. Molecules. 2009;14(12):5144-5164.

9. Kusumaningrum YN. Aktivitas antibakteri ekstrak kulit rambutan (nephelium lappaceum) terhadap staphylococcus aureus & escherichia coli. Dep Biokimia Fak Mat dan Ilmu Pengetah Alam Institus Pertan Bogor. 2012.

10. Nuria MC, Faizatun A. Uji Aktivitas Antibakteri Ekstrak Etanol Daun Jarak Pagar (Jatropha Curcas L) Terhadap Bakteri Staphylococcus aureus ATCC 25923, Escherichia coli ATCC 25922, dan Salmonella typhi ATCC 1408. Medicago. 2009;5(2).

11. Cowan MM. Plant products as antimicrobial agents. Clin Microbiol Rev. 1999;12(4):564-582.

12. Campbell NA, Mitchell LG, Reece JB, Bishop J. Biology: Concepts & Connections. Benjamin Cummings Menlo Park, Calif.; 1997.

13. Fajaryanti N, Kurniawati NI. EFEK antidiare Infusa daun ciplukan (physalis angulata linn) pada mencit jantan putih (mus musculus) yang terpapar oleum ricini. J Farmasetis. 2018;7(1):19-22.

14. Sarayar H, Liwang F. Pencegahan dan penatalaksanaan terkini penyakit travelers diarrhea untuk wisatawan yang berkunjung ke indonesia. Intisari Sains Medis. 2012;1(1):36-40.