Original Research Article

Test the effectiveness of antibacterial effect of the skin of the pomegranate fruit (*Punica granatum*) extract against the growth of *Escherichia coli in vitro*

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ABSTRACT

**Background:** Pomegranate (*Punica granatum*) is one of the traditional medicines that contains antibacterial compounds that are effective against bacterial growth. Its chemical content includes saponins, tannins and flavonoids. *Escherichia coli* is a gram-negative bacterium which is a normal flora germ found in the human large intestine. These bacteria are pathogenic when they are outside the intestine and produce enterotoxins in epithelial cells which cause diarrhea. The purpose of this study was to determine the antibacterial effect of pomegranate skin extract on the growth of *Escherichia coli*.

**Methods:** This study used an experimental design study with a complete randomized design study divided into 5 groups namely groups 1 (15 ug/ml), 2 (20 ug/ml), 3 (25 ug/ml), positive control (ciprofloxacin), negative control. Making pomegranate peel extract was done by maceration method then rotary, after that the effectiveness of pomegranate extract extracted by the diffusion method was tested using Anova one-way test.

**Results:** The results showed that the extract of pomegranate peel showed that it was able to inhibit the growth of *Escherichia coli* with a ratio of constants (15 ug/ml, 20 ug/ml, 25 ug/ml with inhibition diameters of 6.7 mm, 6.7 mm, 6.7 mm, while for positive control with ciprofloxacin showed bacterial resistance to antibiotics.

**Conclusions:** Statistically, pomegranate skin extract has an antibacterial power which is meaningful with p 0.005. Pomegranate rind extract has antibacterial activity against *Escherichia coli* medium because inhibition zone is 5-10 mm.

**Keywords:** Antibacterial, *Escherichia coli*, Pomegranate

INTRODUCTION

Diarrhea is a major cause of morbidity and mortality in children under five, estimated at about 1.3 million deaths of children under five years of age occur all over the world. The prevalence of high incidence of diarrhea in Indonesia in the year 2018 occurs in young toddlers (age 1-5 years), namely by 6.7%. Morbidity and mortality of diarrhea in Indonesia is still high.

Based on a survey of morbidity carried out by the Ministry of Health of the year 2010 s/d 2018 noticeable trend of increased incidence of diarrhea. Percentage of the morbidity of diarrhea in the year 2010 is 37.4%, then increased to 42.3% in 2012, and decreased by 41.1% in year 2013. The percentage of cases of diarrhea in children under five in the Province of West Sumatra in the year 2018 by 31,400 cases (department of health of West Sumatra Province, 2018). Diarrheal diseases still occupy the order of 10 common diseases in the City Padang.

*Escherichia coli* is often the cause of infection of the urinary tract, biliary tract and other places in the
abdominal cavity. *Escherichia coli* is the cause of diarrhea and tract infections kemih. 5.  

Each active substance has a different mechanism as an antibacterial. Previous research has shown that the methanol extract of pomegranate peel has been shown to have activity against the bacteria *Shigella dysenteriae*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *E. coli*. 11. The purpose of this study was to determine the antibacterial effect of pomegranate skin extract on the growth of *E. coli*.

**METHODS**

This study used an experimental design study with a complete randomized design study divided into 5 groups, group 1 (25 ug/ml), 2 (20 ug/ml), 3 (15 ug/ml), positive control (Ciprofloxaatin 500 mg), negative control (aquades). Ciprofloxaatin 500 mg dosage form was suspension powder that was liquefied into 1 cc of sterile aquades. After that the antibiotic disk was dissolved. Making pomegranate peel extract was done by maceration method then rotary, after that the testing of the effectiveness of pomegranate peel extract was done by diffusion method by using ANOVA one-way test.

This research was conducted at the regional health laboratory of West Sumatra Province. Held on 4th November - 7th December 2019. With inclusion criteria ripe pomegranate skin. Making pomegranate rind extract was done by maceration method then rotary, after that the testing of the effectiveness of pomegranate peel extract was carried out by diffusion method.

**RESULTS**

The results of the research can be seen in Table 1 based on the calculation that has been done. Based on the calculation that has been done, the average value of the diameter of the inhibition zones in group 1 at 6.7 mm, group 2 of 6.7 mm, group 3 by 7.7 mm, and the negative control group and positive control do not have the power resistor because the average value of the diameter of the inhibitory zone of 0.00 mm.

Because of the usefulness of this, pomegranate has a lot of be used as one of the alternative treatment traditional. 12

**Table 1: Inhibition zone diameter of pomegranate rind extract (Punica granatum) against Escherichia coli bacteria by diffusion method.**

| Treatment | Deuteronomy | 1 | 2 | 3 | Average (D*) |
|-----------|-------------|---|---|---|--------------|
| Positive control | 0 | 0 | 0 | 0 | 0 |
| Negative control | 0 | 0 | 0 | 0 | 0 |
| 1 (15 ug/ml) | 6 mm | 7 mm | 7 mm | 6.7 mm |
| 2 (20 ug/ml) | 6 mm | 7 mm | 7 mm | 6.7 mm |
| 3 (25 ug/ml) | 7mm | 8 mm | 8 mm | 7.7 mm |

**DISCUSSION**

The results of the study showed that the average value of the diameter of the inhibition zone of one group of 6.7 mm, a group of two 6.7 mm, a group of three 7.7 mm, negative control group and positive not have a zone of inhibition. In the positive control group was given treatment with the antibiotic ciprofloxaacin, the zone of

**Plants that can be used as a herbal medicine d acts as an antibacterial is the pomegranate (Punica granatum L.). 9**

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inhibition is not formed. It is shown that ciprofloxacin is not likely to provide the effect of the inhibition at low concentrations. So that confirms that the results of the antibiotic ciprofloxacin resistance to bacteria *E. coli*. Antibiotics group florokuinolon the most widely used for the treatment of infections is ciprofloxacin, especially that caused by gram-negative bacteria, especially *E. coli*. Resistance *E. coli* to the antibiotic ciprofloxacin is generally caused by chromosomal mutations in the genes gyr A and par C. However, recent research shows that resistance to a low level can also be mediated by plasmids through the acquisition hen qnr-mediated by plasmids pMG252. Increased resistance of *E. coli* to the antibiotic ciprofloxacin has been widely reported. Other studies reported as much as a 20,65% isolates of *E. coli* resistant against ciprofloxacin among the 155 isolates clinic *E. coli* in Pakistan. A specimen of urine patients with UTI in RSUD Abdoel Moeloek (RSUDAM) Lampung Province and obtained 30 isolates positive for *E. coli*. Research in Makassar found 48% isolates of *E. coli* resistant ciprofloxacin among the 39 isolates positive for *E. coli*. According to the WHO (2012), inaccuracy as well as not rational the use of antibiotics is the cause of most of the main spread of resistant microorganisms. So, a drug effective for the treatment, then it should reach the place of its activity in the body with accuracy and a sufficient amount to produce a concentration of effective. Antibiotics will experience transportation is dependent with the power process to plasma proteins. The form that is not bound by such a protein which is pharmacologically active, have the ability as antibakteri. Other mechanisms that cause resistance is decreased accumulation of the drug in the cells by the increase in the pump efflux to native and decreased membrane outer porins. Some species of *Enterobacteriaceae*, including bacteria *E. coli* have a chromosomal native pump efflux AcrAB-ToIC which belongs to the families RND (resistance-nodulasi division).

The criterion of strength antibacterial power is divided into negative control group did not have a zone of inhibition in the inhibitory zone of 5-10 mm. The criterion of strength antibacterial power is divided into three, name antibacterial compounds are divided into three, namely inhibiting the synthesis of nucleic acids, inhibits the function of cell membranes, and inhibit the metabolism energy.

The mechanism in inhibiting the synthesis of nucleic acids is to inhibit the formation of DNA and RNA through the ring A and B which play a role in hydrogen bonding. This leads to the buildup of bases of nucleic acid, and the occurrence of damage to the permeability of the bacterial cell wall, lysosomes, as well as mikrosom.

The mechanism in inhibiting the function of cell membranes is by forming complex compounds with proteins of the extracellular and dissolved which cause damage to bacterial cell membranes and followed by a discharge of compound is intracellular. While the mechanism of flavonoids in inhibiting energy metabolism is to inhibit the cytochrome C reductase and inhibits the use of oxygen on the bacteria. Whereas the energy needed bacteria in the conduct of the biosynthesis of macromolecules. Alkaloids are nitrogen compounds heterocyclic containing at least one nitrogen atom and alkaline Cluster bases will react with the acidic compounds that exist in bacterial cells such as DNA, which is the main constituent of the cell nucleus. With the disruption of the DNA, then the synthesis of proteins and nucleic acids in the cells will disturbed.

**CONCLUSION**

Statistically the ethanol extract of pomegranate peel has antibacterial power that meaningful with p 0.005. The ethanol extract of pomegranate peel has antibacterial activity against the bacteria *Escherichia coli* was due to the inhibitory zone of 5-10 mm.

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**Conflict of interest:** None declared

**Ethical approval:** The study was approved by the Institutional Ethics Committee

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