

Antimicrobial Activity of Lemon (*Citrus limon*) Peel Extract Against *Escherichia coli*

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Abstract

In developing country herbal medicine used widely to treat infectious disease. The *Citrus limon* peel rich in nutrient such as flavonoid and essential oil that can used for antimicrobial and anticancer activity. The *Citrus limon* peel extracted with ethanol 96% using maceration extraction and then dissolved with DMSO. The antimicrobial activity tested against *Escherichia coli* using disc diffusion assay. The present study shows the antimicrobial activity of the ethanolic extract of the *Citrus limon* peel of 25%, 50%, 75% and 100% had a strong antimicrobial activity against *Escherichia coli* were 15,03 mm; 16,17 mm; 15,83 mm; 18,77mm of the average inhibition zone, respectively. The essential oil can disrupt the bacterial membrane and damage the lipids and proteins layer. The flavonoid can inhibit specific enzymes and scavenge free radical. It concluded that lemon (*Citrus limon*) peel extract have a high potential on antimicrobial activity against the *Escherichia coli*.

Keywords: *Citrus limon* peel; *Escherichia coli*; antimicrobial.

1. Introduction

Medicinal plants have been used for therapeutic goal and around 80% of the population in this world uses herbal medicines to treat disease especially for infectious diseases. This herbal medicine widely used in developing country than developed country [1]. The problem of microbial resistance to the available antibiotics can made the traditional medicine can be used as a therapeutic natural agent [2]. Lemon (*Citrus limon*) is an important medicinal plant that was cultivated for its alkaloids, which have anticancer activity and antibacterial potency [3].

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Besides its anticancer activity and antibacterial potency, lemon (*Citrus limon*) can used for skin care, weight loss, good digestion, relief from constipation, eye care, and treatment of scurvy, piles, peptic ulcer, respiratory disorders, gout, gums, and urinary disorders [4]. Lemon (*Citrus limon*) peel just become garbage and make pollution. Lemon (*Citrus limon*) peel rich in nutrient that can used as drugs and as dietary supplements too. These products have less side effects, easily to find and affordable cost. Since there is an increase in the number of antibiotic resistance with some bacteria, there is always a search of an alternative drug [5,6]. The antimicrobial and antioxidant activity on lemon being associated with flavonoids and essential oil [7]. Flavonoids such as hesperidine and naringin are beneficial nutrients that enhance the activity of white blood cells and boost the body's defenses [7,8,9]. Essential oil includes monoterpenes, sesquiterpenes, alcohols, aldehydes, ketones and esters. Monoterpenes had a higher antimicrobial activity than did hydrocarbons. Carvone and limonene were active against wide spectrum of antifungal and antimicrobial activity [8,10]. The aim of the present study was to investigate the antimicrobial activity of the ethanolic lemon (*Citrus limon*) peel extract against *Escherichia coli*.

2. Experimental Section

Materials

Materials used in this study are lemon (*Citrus limon*) peel, ethanol 96%, Dimethylsulphoxide (DMSO), nutrient broth (NB) and nutrient agar (NA).

Instrumentation

Instrument used in this study are autoclave, laminar air flow cabinet, petri dishes, Erlenmeyer, incubator, beaker glass, blank discs, micro pipet.

Procedure

2.1. Preparation of lemon (*Citrus limon*) peel extract

Lemons (*Citrus limon*) bought at local market in Medan, Indonesia. Lemons (*Citrus limon*) washed on the flowing water and peeled them. Dry the lemons (*Citrus limon*) peel under the shade in room temperature. The peel extracted with ethanol 96% using maceration extraction.

2.2. Preparation of extract stock solution

The stock solution of lemon (*Citrus limon*) peel extract was dissolving in a suitable volume of Dimethyl sulfoxide (DMSO) to get a preparation of extract. The concentration of extract can be calculated used the formula:

$$\text{concentration (\%)} = \frac{\text{volume of extract}}{\text{volume of solvent}} \times 100\%$$

2.3. Antimicrobial activity of lemon (*Citrus limon*) peel extract against *Escherichia coli*

Antimicrobial activity of *Escherichia coli* was investigated using disc diffusion assay. *Escherichia coli* was transferred into 13 mL sterile nutrient broth and incubated at 37°C for 18-24 hours. After incubation, put in 0,1 mL *Escherichia coli* into the petri dish and add in 15 mL nutrient agar into the petri dish, mix them and wait until the nutrient agar solidified. The blank discs (diameter 5 mm) were dipped into the extract stock solution and then plated on the surface of nutrient agar. After 24 hours incubation, the inhibition zones were determined using calipers.

3. Result and Discussion

3.1. Lemon (*Citrus limon*) peel extract

The lemon (*Citrus limon*) peel extracted with ethanol 96% using maceration extractor are shown on Figure 1.



Figure 1: Lemon (*Citrus limon*) peel extract

3.2. Antimicrobial activity of lemon (*Citrus limon*) peel extract

The lemon (*Citrus limon*) peel extract were determined for their antimicrobial activity against *Escherichia coli* using disc diffusion agar method. The evaluation of antimicrobial activity was conducted triplicate. Diameter of inhibition zone of lemon (*Citrus limon*) peel extract were shown in table 1. The strength of the antimicrobial inhibitory is shown in the bar chart on figure 2.

Table 1: inhibition zone of lemon (*Citrus limon*) peel extract against *Escherichia coli*

Name of the compounds and their concentration	Antibacterial activity diameter of inhibition zone against <i>Escherichia coli</i> (mm)			
	I	II	III	Mean
Peel extract (25%)	15,10	15,80	14,20	15,03
Peel extract (50%)	16,10	16,30	16,10	16,17
Peel extract (75%)	14,20	16,10	17,20	15,83
Peel extract (100%)	18,80	19,20	18,30	18,77

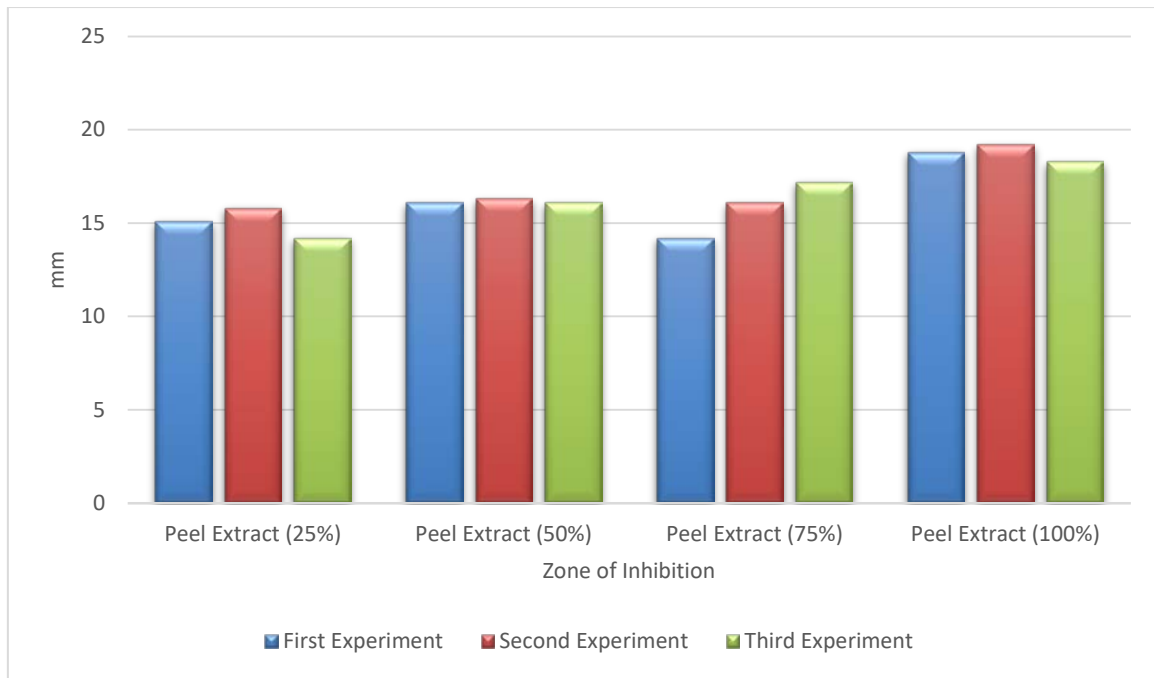


Figure 2: Zone of inhibition according to peel extract concentration

Table 1 showed that compounds of lemon (*Citrus limon*) peel extract were effective to inhibit *Escherichia coli*, where in peel extract (100%) showed the strongest antimicrobial activity. The antimicrobial activity of lemon (*Citrus limon*) peel extract against *Escherichia coli* showed in figure 3.

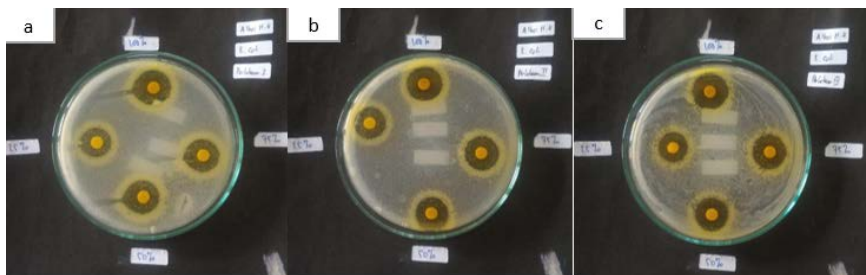


Figure 3: The antimicrobial activity of lemon (*Citrus limon*) peel extract against *Escherichia coli*; (a) first experiment, (b) second experiment, (c) third experiment

Figure 3 showed the result of inhibition zone on 100% concentration greater than the other concentration. Based on this result, the strength on lemon (*Citrus limon*) peel extract on concentration 100% gave 18,77 mm of the average inhibition zone. The similar result was reported by Tumane and his colleagues [11], which reported that diameter from zone of inhibition against *Escherichia coli* from lemon (*Citrus medica L.*) peel extract using soxhelt extraction was 12 mm. Mansour and his colleagues [12] reported that diameter from zone of inhibition against *Escherichia coli* from Algerian Lemon (*Citrus limon L.*) peel extract was 20 mm.

The essential oil includes limonene and γ -terpinene related to antimicrobial effect against *Escherichia coli* can disrupt of the bacterial membrane and inhibit the respiration and ion transport processes [13]. Therefore, the

permeability of the membrane increased. Essential oils pass through the cell wall and cytoplasmic membrane may also coagulate in the cytoplasm and damage the lipids and proteins layers [14]. Flavonoid has an antibacterial, antimicrobial, antioxidant and antiinflammatory activities, which proven ability to inhibit specific enzymes, to stimulate some hormones and neurotransmitters, and to scavenge free radicals [15].

4. Conclusion

From the result it can be concluded that lemon (*Citrus limon*) peel extract have a high potential on antimicrobial activity against the *Escherichia coli*.

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