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The Synergistic Antibacterial Effects of Cinnamomum verum and Cymbopogon citrates on Urinary Tract Infections causing pathogens Escherichia coli and Pseudomonas aeruginosa

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Abstract: Escherichia coli (E. coli) and Pseudomonas aeruginosa (P. aeruginosa) have been one of the few main pathogens that lead to urinary tract infection. The study was conducted because there have been an increase in antibiotic resistance pattern in the world. Ethanol and aqueous extract of medicinal plant, cinnamon, Cinnamomum verum and lemongrass, Cymbopogon citrates were prepared to test its synergistic antibacterial effect against . The test was then preceded with the Minimum Inhibitory Concentration (MIC) using the 96 well microtitreplate. The Minimum Bacterial Concentration (MBC) were then attained after the the MIC result were observed. The MIC of E.coli was at 6.25 µg/mL for the synergy ethanolic extract between Cinnamomum verum and Cymbopogon citratus. The aqueous extract did not visualize any growth inhibition on the bacteria E.coli. Both ethanolic and aqueous extract also had no inhibitory effect on the P. aeruginosa. The synergistic extract of Cinnamomum verum and Cymbopogon citratus was effective against E. coli. Therefore, it was concluded that the effect of the extract was effective on E. coli and it was almost as effective as the antibiotics that were usually prescribed for the treatment of urinary tract infections.

Key words: Synergy, Cinnamomum verum, Cymbopogon citratus, Escherichia coli, Pseudomonas aeruginosa.

INTRODUCTION

Antibiotic treatment has been widely used for the treatment of urinary tract infection, this has also contributed to the rise of antibiotic resistance in the population. From 2000 to 2010, the antimicrobial resistance of urinary tract pathogen, Escherichia coli towards the antibitiotic ciprofloxacin and trimethoprim and sulfamethoxazole have increased rapidly. Cinnamon is one of the most well known common spices that can be found all over the world. It has also been used both as cooking material and medicine over thousands of years. Cinnamon comes from the family of Lauracea. The cinnamon is derived from the inner bark of trees from the genus Cinnamomum. The other plant that was used in this synergistic antibacterial study was lemongrass. The plant is scientifically known as Cymbopogon citratus (C.citratus). One of the recent studies have stated that the ethanolic extracts of the leaves of lemongrass showed potential antibacterial property against Staphylococcus aureus.
OBJECTIVE

The main aim of this study was to investigate the synergistic antibacterial effect of cinnamon, *Cinnamomum verum* and lemongrass, *Cymbopogon citratus* against *Escherichia coli* and *Pseudomonas aeruginosa*.

METHODOLOGY

| Plant Extraction | Cut the plant into small pieces.  
|                  | soaked briefly with distilled water before blotting the excess water with tissue paper.  
|                  | placed in a clean steel tray and were set in the hot air oven at 40°C.  
|                  | left to dry in the oven.  
|                  | the plants had to be finely grinded using a dry blender to obtain the powdered formed.  
|                  | about 200g of fine powder of both plants were weighed and were soaked in 600ml of 95% of ethanol and water.  
|                  | soaked for 3 days.  
|                  | The aqueous extract were placed in the hot air oven at 40°C while the ethanolic extracts were be concentrated using the rotary evaporator also at 40°C.  
|                  | Finally, the extracts were chilled in the refrigerator until used at 4°C.  

| Bacterial Strains (Inoculum Preparation) | Cultured the pure culture-24 hours at 37° C.  
|                                          | stored at 4° C to maintain its growth.  
|                                          | The inoculums were prepared by growing the sub culturing the bacteria that had been made in Mueller Hinton broth at the temperature of 37° C, overnight.  

| Antibiotic Susceptibility Test | Using the disk diffusion method (Kirby Baeur test).  
|                               | bacteria inoculum was prepared by inoculating a single colony of the bacteria *E. coli* and *P. aeruginosa* into the Mueller Hinton broth (incubated at 37°C overnight).  
|                               | the paper disk which contained single concentration of each antimicrobial agent will be set on the surface of the agar that has been inoculated with the bacteria.  
|                               | blank disc that had been soaked in the extract of the *C. verumand C. citratus* were also placed on the agar for testing.  
|                               | Three different concentration of tested plant extract were prepared (20 mg/mL, 50 mg/mL and 100 mg/mL). For *E. coli*, the positive control that was used for the experiment was |
ciprofloxacin. As for *P. aeruginosa*, the positive control was gentamicin).

- The negative control used for both organism was distilled water for aqueous extract, 10% DMSO for ethanolic extract.
- The extract of the cinnamon and lemongrass antibacterial effect would be compared with the commonly used antibiotic.
- the agar plate was leaved to be incubated overnight at 37°C and measurement of inhibition zone was taken the following day.

**FINDINGS**

As stated by Ochei et al. in the book of Medical Laboratory Science, Theory and Practice year 2000, there are a few factors that can affect this antimicrobial susceptibility test. The first is the depth of the medium. The depth of the media that is being used may affect the result as it can interact and disturb the concentration gradient of the diffusing antibacterial or antimicrobial agent. Thicker agar medium will give out smaller zone of inhibition while thinner agar medium will produce a larger zone. In order to standardize, it is advisable to pour 25 ml of medium in agar plate which will produce an agar with the depth of 4mm. Next factor that can interfere with the test is the atmosphere of the incubator. The incubation is recommended to be carried out in incubator with ambient air. If the incubation is carried out, for an example in a carbon dioxide incubator, it may result in decrease pH of the agar, thus affecting the action of some drugs.

**CONCLUSION**

The synergistic ethanolic extract of *Cinnamomum verum* and *Cymbopogon citratus* was effective against one of the bacteria which were *E. coli*. The result that were obtained had also shown that effect of the extracts were almost as effective as the antibiotics that were usually prescribed for the treatment of urinary tract infections. To ensure the safety of the extract, the extract can also be tested against the normal flora of the urinary tract. Toxicity test of the extract can also be done. The extract can also be tested on extended spectrum beta lactamase (ESBL) bacteria, in order to know more about its effect on Gram negative bacteria.

**SELECTIVE REFERENCES**
