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ANTIMICROBIAL ACTIVITIY OF SOME PLANTS EXTRACTS AGAINS MRSA

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Abstract

This study is applied to determine the effectiveness of some plants extracts against Methicillin-resistant *Staphylococcus aureus* MRSA **by** in vitro disc diffusion method to find potential antimicrobial effect of some plant of some plants that used and to cure infectious disease in herbal and traditional medicine in Saudi Arabia. MRSA is specific strain of *Staphylococcus aureus*, it causes the same disease$^{s'}$ symptoms seen in other staph infections, but the MRSA strain is resistant to treatment with commonly used antibiotics. *krameriaceae* (Khawajoawa) *Melaleuca alternifolia* (Tea tree oil), and *Acacia nilotica* (Garad), are plants, believed by herbalists to have antimicrobial effect. These plants have been tested in the present study to investigate their in vitro potential antimicrobial effects against MRSA. First the plants were extracted, the polar compounds of *Rhatany krameriaceae* was extracted with ethanol Show narrow inhibition zone of 19mm, the polar compounds of *Acasia nilotica* was extracted by distilled water Show

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inhibition zone of 24mm, The commercial product of tea tree oil extract use as non-polar compounds of *Melaleuca alternifolia* (Tea tree oil), show large inhibition zone 48mm, the extracts were tested by the filter paper disc method.

Introduction and **Review**

Methicillin-resistant *Staphylococcus aureus* is a gram positive bacterium that is genetically different from other strains of *Staphylococcus aureus* . MRSA strain is resistant to treatment with commonly used antibiotics in contrast to the remainder of the *Staphylococcus aureus*groupwhich are referred to as methicillin sensitive *Staphylococcus aureus*(MSSA). MRSA responsible for several difficulttotreat infections in humansand the symptoms it causes are the same as the symptoms seen in other *staph* infections of the skin (Coella R et al 1997)

Staphylococcus aureus has shown an ability to resist antibiotics during the last 40 years ,about 90% of strains of *staph aureus* found in the hospitals are now resistant. Resistance to penicillin depends on production of the enzyme penicillinase (Jose M. and Cesar 2016). MRSA is any strain of *S. aureus* that has developed, through horizontal gene transfer and natural selection, multiple drug resistance to beta-lactam antibiotics. β-lactam antibiotics are a broad spectrum group which includes some penams – penicillin derivatives such as methicillin and oxacillin, and cephems such as the cephalosporins (Gurusamy et al 2013) MRSA strains are resistant to all β-lactam agents and often to other agents such as the aminoglycosides and fluoroquinolones(Jose M. and Cesar 2016). The MRSA considered as bacteria resistant to antibiotics so it's more difficult to treat but studies have shown that are some therapeutic plant that used as antibacterial, parasite and fungus and it help in MRSA cure
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Rhatany krameriaceae have Antibacterial, anti-oxidant and astringent medicine and used in herbal medicine and traditional medical treatments(Asian Pac J Trop Biomed. 2012)

The essential oil of Melaleucaalternifolia, commonly known as tea tree oil, has a long history of use as a topical antiseptic (Markham 1999)

-tea tree oil is effective *against E.coli and staph* infection when combined with eucalyptus , one of recommendation for helping fight infection found in chest colds (Gurusamy et al 2013) The roots make a mucilaginous tea that is both antibacterial and anti-inflammatory. It helps soothe mucous membranes from the mouth through to the anus, reducing inflammation and attacking microbial infections.

Materials and methods
Tea tree oil for this study is brought from one of American company approved by the Food and Drug Administration. This product is organic and have a concentration

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about100%, under sterilization conditions to be used properly, we putting this product in autoclave with the discs to avoided any contamination. For *krameria triandraon*

Roat were finely powdered using an electric grinder and extracted by ethyl alcohol concentration of 95% was carried out at room temperature with repeated operation three times. Then the filtration process was done to remove the residue of the plant powde .The extract was then concentrated to remove the alcohol used in extraction process using a rotary steamer under low pressure. The extract in its current form contains a small amount of alcohol which is required to transfer the extract from the device to the flask.

Acacia nilotica air dried seeds were finely powdered using an electric grinder. For conventional extraction 5 g of powdered plant material was mixed with 10 ml of distilled water in a round bottom flask, it was then autoclaved at 121 °C and at 15 lbs pressure and stored at 4 °C (Jigna and Sumitra, 2007).

Preparation of sensitivity discs were done in the laboratory. Whatman,s No 1 filter paper were used. These were obtained by punching the filter paper with a paper punch (6mm diameter). The disc were sterilized by autoclaving at 1210c for 15 minutes Filter Paper Disks is prepared by cutting the filter paper at approximately 6 mm in diameter then it must be sterilized this is done by putting the disks in are placed in a Petri dish and sterilized in a hot air oven (Brander and Bugh, 1977).Then the Filter Paper Discs soaked in tested plant extractions until were completely absorbed extractions. Then the discs are removed and put in sterile Petri dish.

Mueller Hinton Agar is used for determination of plat extraction susceptibility test

Preparation of inoculums (turbidity test) :

1) Using a sterile inoculating loop or needle, touch four or five isolated colonies of the organism to be tested.

2) the organism is suspended in 2 ml of sterile saline.

3) Vortex the saline tube to create a smooth suspension.
4) Then we put the tube contain MRSA suspension in the turbidity machine . By international convention we adjust the turbidity of our culture against the standard 0.5 MacFarland to be able to compare between microorganisms, antibiotics according to the specifications of the NCCLS. (U. Eigner et al 2005)

5) Use this suspension within 15 minutes of Inoculation of the MH plate: (all steps done according the aseptic technique**)(** Jan Hudzicki 2009)

The bacteria cultured by sterile swab which dip into tube contain bacterial suspension after that swab rotated against the side of the tube (above the fluid level) using firm pressure, to remove excess fluid. Then swab should not be dripping wet Inoculate the

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dried surface of a MH agar plate by streaking the swab three times over the entire agar surface; rotate the plate approximately 60 degrees each time to ensure an even distribution of the inoculum after that sit at room temperature at least 3 to 5 minutes, for the surface of the agar plate to dry before add the Filter Paper Disks that contain the reagent to be tested by flamed and cooled forceps

the disc was Press gently into the agar with the help of the forceps, The space between the discs must not be narrower than 24 mm, and the distance to the edge no less than 1 cm., the plate incubated at 37°C 24 hours. After that inhibition zones are be measured

RESULT and Discussion

In the present study three plants parts namely *krameriaceae* (Khawajoawa) *Melaleuca alternifolia* (Tea tree oil), *Acacia nilotica* (Garad), and *Boswellia carterii* Frankincense (also known as *olibanum*) which are believed amongst herbal therapists as antimicrobial agents, were examined. Tests were made to find their possible in vitro effects by observing the inhibition of growth of MRSA.

Three different methods were use to extract the chosen plants parts, ethanol used to extract polar compounds of *krameriaceae* (Khawajoawa), and fatty compounds (nonpolar) of *Melaleuca alternifolia* (Tea tree oil), used in oil form, *Acacia nilotica* (Garad) wias been extracted by distilled . The plant extracts were tested using discs method. This method was found to be suitable to screening potential antimicrobial effects of plants extraction Because the results of the extract efficacy is very easy to read, the result had been taken by measuring the inhibition zone by transparent ruler in millimeter The results of Tea tree oil extract against MRSA strains using disc method are shown in Table (1). Figures (1). bacteria was inhibited with large inhibition zone 48mm. There was good result with *A. nilotica* against tested strains using disc method shown in Table (1). Figures (2). Show inhibition zone of 24mm. But *krameria triandraon* (Khawajoawa) Show inhibition zone of 19mm inTable (1) Figures (3).

Figure 1: inhibition zone of tea tree oil

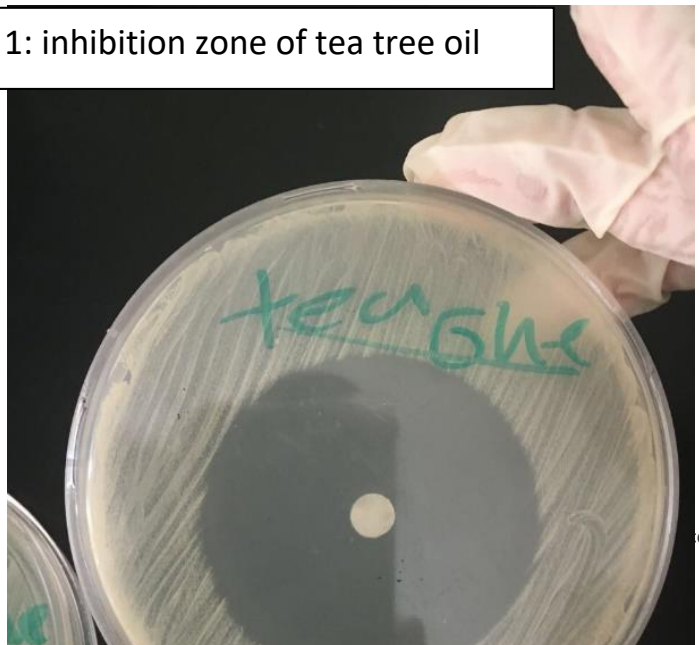

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Figure2: inhibition zone of *Acacia nilotica*:
Show inhibition zone of 24mm

Krameria triandraon:show inhibition zone of 19mm

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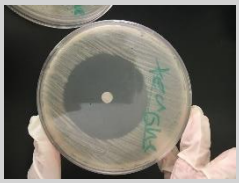
Table (1)

plant	**Inhibition zone**	**Zone diameter**	**Photo**
TEA TREE OIL	There is	48mm	
Krameria triandraon	There is	19mm	
Acacia nilotica	There is	24mm	

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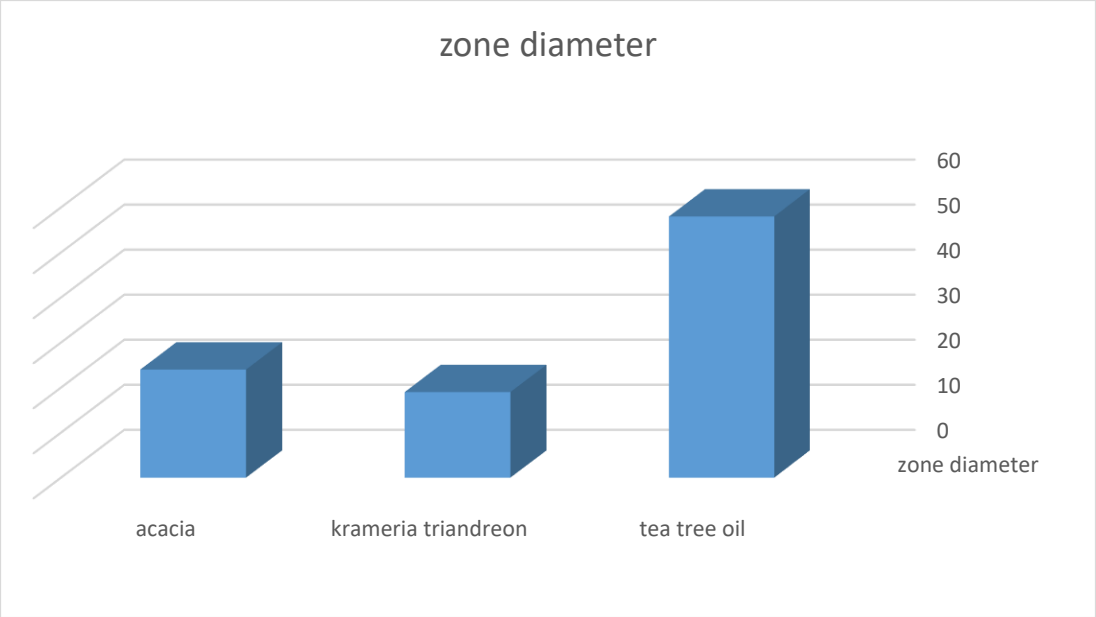

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