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Effect of Olibanum Essential Oil on Bacterial Plasmid

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Abstract: *Essential oils are concentrated natural extracts derived from plants, which were proved to be good sources of bioactive compounds with antimicrobial properties. Olibanum essential oil has become increasingly popular in skin care. The current study exhibited the effect of this essential oil on curing resistant plasmid of Staphylococcus aureus isolated from burned skin. The oil was extracted by using Clevenger apparatus.Alkaline lysismethod was done to determine the antiplasmid activity. Loss of plasmid band was confirmed by agarose gel electrophoresis. The curing efficiency of olibanum essential oil was found to be 100% in all concentrations (0.5%, 1%, 3%) as compared with control. In conclusion, the essential oil of Boswellia resin may be considered as a natural source for antimicrobial and antiplasmid agents and could be used for application in pharmaceutical industries.*

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I. Introduction

Medicinal plants consider as rich resources of ingredients which can be used in drug development and synthesis.1.Boswellia species are economically and ecologically important plants.2.This genus has 25 speciesbelong to the Burseraceae family, the common use are (B.carteriandB.papyrifera) in Ethiopia, B.serrata in india and B.sacra in southern Arabia.3. Boswellia species have aromatic gum resin obtain from trees by made incisions in the trunks of the trees to produce exuded gum that has the appearance of a milk-like resin. Upon exposure to air, the resin hardens into orange-brown gum known as frankincense. Frankincense oil is one of the most commonly used oil in aromatherapy and it is also used in incense, as a fixative in perfumes.4.It is also used as a raw material in several industries such as pharmacology, food, beverage, flavoring, cosmetics, detergents, creams , paints, adhesive and dye manufacturing.2.Different researches revealed that this essential has the ability to act as anti-cancer.5, especially against skin cancer6, as well as to its importance as antioxidant and antimicrobial.7. Staphylococcus is a genus of Gram-positive bacteria .Most are harmless and reside normally on the skin and mucous membranes of humans and other organisms.S.aureus is responsible for a broad spectrum of human and animal diseases, not just skin infections but severe diseases such as pneumonia and endocarditis.8.Infections by some species of staphylococci are difficult to treat because of frequency of multiple antibiotic resistant strains The genes that are responsible for antibiotic resistance in S. aureus are usually found on the plasmids or some other structures similar to plasmids. Plasmid profiles have been useful in the detected sensitivity of bacteria and in tracing antibiotic resistance.9. Hence, there is continuing need for newer and effective plasmid curing agents who are effective and less toxic that could cure the plasmid encoded antibiotic resistance in broad spectrum of bacteria.So this study was done to investigate the activity of olibanum oil as a natural antiplasmid agent.

II. Material And Methods

Plant material and extraction

Olibanum (Resin) was purchased from the local market.100 g from grinded resin were submitted to steam distillation using the Clevenger apparatus for 4 hours.[10]. Oil was stored in a separated marked glass vials in a refrigerator until required**.**

Test microorganism

The microorganism used in this study include gram positive bacteria *Staphylococcus aureus.* The microorganism is a clinical isolated fromburned skin .

Alkaline lysis mini prep procedure

Plasmid was extracted by using the following alkaline lysis method.[11.]

1. Inoculate a 5 ml culture of LB/amp (50-100 µg/ml) with a single bacterial colony. Place tube in 37°C shaker overnight.

2. Fill an Eppendorf tube with approximately 1.5 ml of the culture and microfuge 1 minute. Remove the supernatant and add another aliquot of culture to the tube. Again, microfuge 1 minute and remove the supernatant. Repeat until the entire 5 ml culture is spun down in one tube.
3. Re-suspend pellet in 100 µl of solution I: 100 ml
Solution I: 25 mMTris pH 8.0, 10 mM EDTA pH 8.0, ml 50 mM Glucose
4- Add 200 µl of Solution II:
1 ml: 50 µl 20% SDS 20 µl ,10 N NaOH 930 µl dH2O
Mix and ice 5 minutes.
5- Add 150 µl Solution III: (3 M K+, 5 M Acetate)
Solution III 100 ml: 5 M KOAc 60 ml glacial acetic acid 11.5 ml dH2O 28.5 ml.
5 M Potassium acetate 100 ml: 49.075 g potassium acetate Fill to 100 ml with dH2O
6-Vortex gently to form small white clumps. Ice 5 minutes
7- Microfuge 5 minutes in cold microfuge.
8- Transfer supernatant to new tube. Add 400 µl of :chloroform. Vortex and microfuge 2 minutes.
9- Transfer aqueous (upper) phase to a new tube. Add 1 ml room temp EtOH. Mix well and stand at room temp for 2 minutes
10- Microfuge 5 minutes in cold microfuge. Pour off EtOH and let pellet dry completely.
11- Place tube at 37°C for 30 minutes.
12- Dissolve the DNA in (50-100)µl TE buffer .

Agarose Gel Preparation and Electrophoresis

Agarose gel was prepared by dissolving 1 g of agarose powder in 100 ml of 1 x TBE buffer and melted then cooled to 50- 60°C. 5 µL of ethidium bromide dye was added to the gel with mixing, the solution was poured into a tray, several wells were carefully made with a comb at one side of the gel. After solidification, the comb was carefully removed and jar was put in the electrophoresis tank.[12]. Each well was loaded with 3 µl of loading dye mixed with 5µl of total DNA, the electrical power was turned on after closing the tank lid then adjusted at 80 volt for 1 hr, the migration of DNA was from cathode (-) to anode (+) poles. Agarose gel tray was placed in gel documentation system for visualization of DNA under ultraviolet light (350 nm) and photographed.[13]**.**

III. Results

Figure 1 shows the presence of plasmid in lane (1) which represented the control but absent in all different concentrations of olibanum essential oil(lanes 2,3,4). This indicate that this bacterial species loss the resistance by the physical loss of plasmid by the activity of the frankincense essential oil which increased with increasing the concentration .

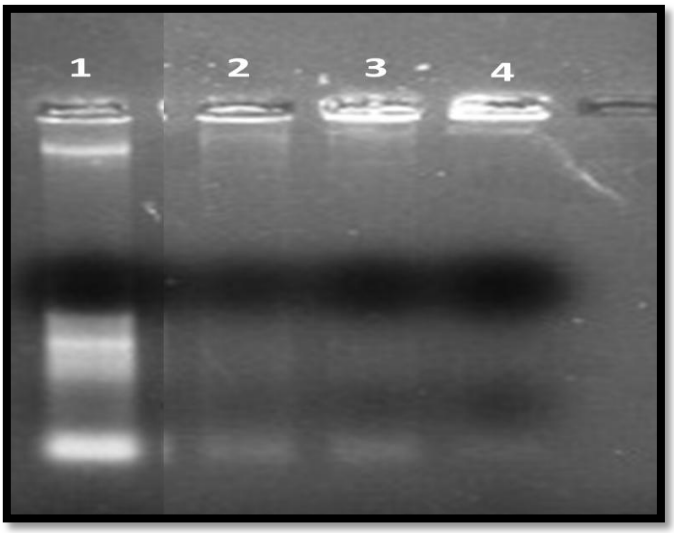

Figure 1:Gel electrophoresis of *S.aureus*plasmid DNA treated with different concentrations of olibanum essential oil : lane (1) control, lane (2) plasmid DNA treated with 0.5 % of olibanum oil (3) plasmid DNA treated with 1% of olibanum oil, lane (4) plasmid DNA treated with 3% of olibanum essential oil.

IV. Discussion

This study suggest that the chemical components of the *Boswellia*essential oil responsible of its activity which were divided according to.[14] , into seven classes in different ratios, including: monoterpene hydrocarbons (6.42%), oxygenated monoterpenes (13.31%), sesquiterpene hydrocarbons (2.09%), oxygenated sesquiterpenes (0.51%), diterpenes hydrocarbons (3.41%), oxygenated diterpenes (21.04%) and other components (53.22%). The major constituents were identified as verticiol (14.48 %), isobutylcyclopentane (12.25%), n-octyl acetate (9.20%) and 9-oxabicyclo-[6.1.0]-non-3-yne (9.12%). On the other hand, the inhibition of plasmid replication at various stages may also be the theoretical basis for the elimination of bacterial virulence in the case of plasmid mediated pathogenicity and antibiotic resistance.[15].

Plant-derived compounds can be effective at curing plasmids in vitro; however, more research is needed to confirm spectrum of activity, identify the active components and to determine any toxicity and in vivo efficacy.

V. Conclusion

Boswellia(olibanum) essential oil may be considered as a natural sourceantiplasmid agents and could be used for application in pharmaceutical industries.

References

[1]. Hassan R. 2012.Medical plant important and use ' Pharmaceuticaanalytica acta.3:10
[2]. Lemenih.M and Teketay.D.2003. Frankincense and myrrh resources of Ethiopia distribution ,production ,opportunities for dry land development and research need .SINET :ETHIOP.J.SCI.;26(1):63-27.
[3]. RameshkumarK.B.2014.Frankincense.https://www.researchgate.net/publication/299436058
[4]. Maloney, G.A. 1997. Gold, frankincense, and myrrh: An introduction to EasternChristian spirituality . New York: C rossroads Pub. Co
[5]. Suhail MM, Wu W, Cao A, Mondalek FG, Fung KM, Shih PT, Fang YT, Woolley C, Young G, Lin HK.2011.Boswellia sacra essential oil induces tumor cell-specific apoptosis and suppresses tumor aggressiveness in cultured human breast cancer cell BMC Complement Altern Med. 15;11:129.
[6]. Fung KM, Suhail MM, McClendon B, Woolley CL, Young DG, Lin HK.2013. Management of basal cell carcinoma of the skin using frankincense (Boswellia sacra) essential oil: A case report. OA Alternative Medicine .1;1(2):14.
[7]. Reddy P. A. , Venkata R. K , B. M. Lepakshi , N. R. Bhumi , V. R. Lebaka .2015. Chemical Profile, Antioxidant and Antimicrobial Activity of Essential Oils from *Boswelliaovalifoliolata* Bal. et. Henry' International Journal of Pharmaceutical and Clinical Research. 7(1): 96-101.
[8]. Dar, J.A., M.A. Thoker, J.A. Khan, A. Ali and M.A. Khan. 2006. Molecular epidemiology of clinical and carrier strains of methicillin resistant *Staphylococcus aureus* (MRSA) in the hospital settings of north India. Ann. Clin. Microbiol. Antimicrobials. 10.1186/1476-0711-5-22
[9]. Sambrook, J., E.F. Fritsch and T. Maniatis, 1989. Small Scale Preparations of Plasmid DNA, in Molecular Cloning: A Laboratory Manual. 2nd Edn., Cold Spring Harbor Laboratory Press, New York, pp: 1.25-1.31.
[10]. European Pharmacopeia, 4th ed.; Council of Europe: Strasbourg Cedex, France (2002) 2.8.12, pp 183-184.
[11]. Brinbiom, H.C. and Doly, J. 1970. A rapid alkaline extraction procedure for screening recombinant plasmid DNA. Nucleic Acids Research 17, 1513-1523.
[12]. Vyas, S.P. and D. V. Kohli .2012. Methods in biotechnology and bioengineering.CBS, 7thed. Publishers & distributors Pvt. Ltd., India.
[13]. Couto, M. C.; Sudre, A. P.; Lima, M. F. and Bromfim, T. C. (2013). Comparison of techniques for DNA extraction and agarose gel staining of DNA fragments using samples of Cryptosporidium. Vet. Med., 58(10): 535-542.
[14]. Mohamed A. A., S. I. Ali, H. F. Kabiel , A. K. Hegazy, M. A. Kord and F. K. EL-Baz.2015. Assessment of Antioxidant and Antimicrobial Activities of Essential Oil and Extracts of *Boswelliacarteri*Resin . International Journal of Pharmacognosy and Phytochemical Research .7(3); 502-509.
[15]. Spengler G., A. Molnár , Z. Schelz , L. Amaral , D. Sharples and J. Molnár.2006. The Mechanism of Plasmid Curing in Bacteria. Current Drug Targets 7(7):823-41 .

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