



Research Article

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Effective of Watery and Alcoholic Extract of Frankincense on the Candida Albicans Fungus

Maha E I Aldory¹, Fawwaz Fadhil Ali^{2*}, Safaa M. Sultan³

¹Ministry of Education Mosul, Iraq,

²Animal Production Division, Technical Institute Mosul, Northern Technical University, Mosul, Iraq,

³Technical Institute Hawija, Northern Technical University, Kirkuk, Iraq.

*Corresponding Author

ABSTRACT

Background. The side effects of some antibiotics against humans, as well as the high prices of these antibiotics, led to dependence on natural antibiotics derived from medicinal plants in order to obtain natural antibiotics without side effects and cheap prices and also available. *Methods.* The frankincense material (that used in this research) is considered to be one of the most common drugs used by humans in relation to many diseases and has no significant side effects. This is due to the fact that this substance contains basilic acids. It is considered the active substance in frankincense. *Objective.* This research was conducted in order to study the effect of the watery and alcoholic extract of frankincense on the fungus of *Candida albicans* as well as for the discovery of the active substance of frankincense and thus to getting a natural antibiotic against this fungus and other organisms which causes many diseases for humans. *Results.* The results showed that the alcoholic extract is more effective than the watery extract. Concentration (200) for the extract of frankincense showed an effect on the *Candida*, and the concentration was increased to (400) and its decreased to (100) was also accompanied by an increase and decrease in the inhibitory activity of the extracts respectively, in comparison to the inhibitory effect of some antibiotics studied on this fungus. *Conclusion.* The Concentration (400) of alcohol extract was superior to the studied fungi.

Key words: *The Frankincense, Candida Albicans. Antibiotic, Watery Extract, Alcohol Extract.*

INTRODUCTION

The development of resistance fungi for treatments, especially the multiple resistance as well as the negative side effects of some of these treatments and the high cost of preparation and manufacturition as well as other reasons all led to consideration to alternative medicine to overcome these problems [1]. Moreover, nowadays medicinal plants field is one of the most important alternatives and get high attention for studying continuously around the world [1].

The frankincense is used in most countries of the world. However, folk medicine found since ancient times called (Frankincense) as well as male, kender, and other species that obtained from the *Boswellia spp* tree of the family Burseraceae [2]. The frankincense tree is of economic importance for many countries in middle east such as Yemen and Oman. Also, frankincense tree found in African countries such as Somalia, Sudan, southern Egypt. Recently, no longer usually about three meters long and with a small or pointed neck or a leaf Ace. The definition of frankincense is a lactic acid extracted by make cuts on the stalks of this tree, where it is dried. They become small pieces in different shapes called the pure section of these cutting the(francincense) [3] As the Figure 1.

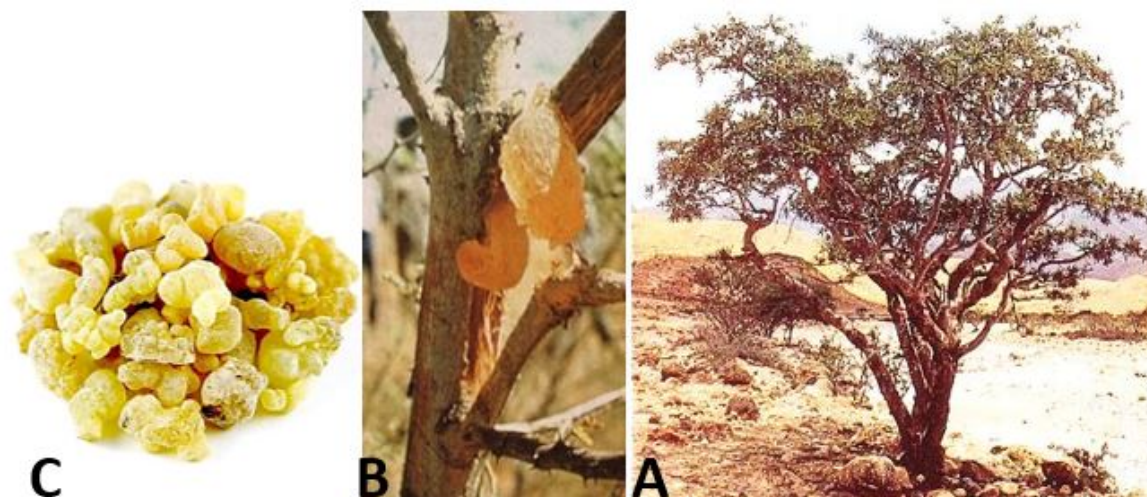


Figure 1. A-The frankincense tree, B- Extract the frankincense from the tree, C- Pure frankincense.

The frankincense is one of the most famous materials that are sold and found in aromatherapy shops and Arab medicinal herbs. In addition, frankincense is used in folk medicine to treat many diseases including tumors, sores, dysentery and chest diseases such as cough, asthma and shortness of breath as well as leprosy as a heart tonic and other non-medical uses most important and most popular [4] which is one of the best materials for the treatment of some chest diseases such as cough and asthma. Beside the strengthening of the bronchial and repellent sputum. It also helps to treat the memory weakness that many people complain about, especially the hardening during the study period, where it mixes with other substances.

On the other hand, the frankincense extract is anti-chronic gingivitis. Also, this herbal medicine may be applied in an easy and safe way to improve gum health, these natural properties of the extract of frankincense offer satisfactory therapeutic properties as well as used in the removal of tooth decay easily, as this is difficult to present at present [5]. There is great importance to frankincense for the nervous system frankincense extract may promote the regeneration of crushed sciatic nerve by improving the sciatic nerve function. However, the exact mechanism has not been clearly defined. Further studies are needed to evaluate the significance of frankincense extract in peripheral nerve regeneration [6]. In addition to the fact that frankincense has a significant impact on the spores of the three types of staphylococcus, and this effect is different from several antibiotics are expensive and not available and disadvantage on the human as opposed to frankincense, which is a natural antiseptic and available and cheap and has no serious side effects on humans [7]. Chemically, frankincense is made up of Resistant materials about (20-36%), volatile oils (4-8%) and colloids acid about (56-65%), and active substances due to colloidal acids, collectively called basilic acids.

On the other hand, Candida is an oval-shaped microorganism which can reproduce rapidly by budding. However, Candida cannot thrive by itself, and almost everyone has intestinal Candida colonies. It is one of many organisms that thrive in our intestinal flora. Under normal circumstances, the candida live in harmony through a delicate balance of bacterial types. A candida infection occurs when a certain set of physiological circumstances allow and encourages its growth. Many people experience sensitivity to yeast that is prone to grow on specific mucous membranes of the body, where the climate is moist and favourable. The vagina, mouth and gastrointestinal tract are areas that frequently grow candida. The climate of the vaginal tract is perfect for candida proliferation, although if biochemical balances are maintained, yeast overgrowth is prevented [8].

Furthermore, Candidiasis is a common infection of the skin, oral cavity and esophagus, gastrointestinal tract, vagina and vascular system of humans. Although most infections occur in patients who are immune compromised or debilitated in some other way, the organism most often responsible for disease, *Candida albicans*, expresses several virulence factors that contribute to pathogenesis. These factors include host recognition biomolecules adhesions.

Additionally, 'phenotypic switching' is accompanied by changes in antigen expression, colony morphology and tissue affinities, *albicans* and several other *Candida* spp. Switching might provide cells with flexibility that results in the adaptation of the organism to the hostile conditions imposed not only by the host but also by the physician treating the infection [9].

MATERIAL AND PROCEDURE :**MATERIAL :**

Frankincense : The Frankincense was obtained from local markets in the city of Kirkuk – Iraq.

Sample: The sample used in this study is the sample of *Candida albicans* fungus obtained from Azadi Hospital in Kirkuk, Iraq. The diagnosis was confirmed by the adoption of vitro and microscopic tests.

The specimen was cultured on the culture media of (the sabouraud dextrose agar), where it was streaking by using the lob, more than one pitre dish of the sample was cultured on the above-mentioned medium to ensure that the fungal growth was not polluted during the culturing process and the dishes were incubated at 37 ° C for 24 hours in a special incubator [10].

Cultural Characteristics :

The developing colonies appeared on the (sabouraud dextrose agar) in the form of white colonies to the cream of color, smooth, and in circular colonies. (Sarah Kidd and his group (2016)) point out that *Candida* spp colonies have such phenotypic characteristics when implanted in the same medium [1, 4] This result is consistent with (Singh and his group (2013)) showing colonies of creamy, smooth and round shape to provide appropriate culturing conditions [11] Figure 2.

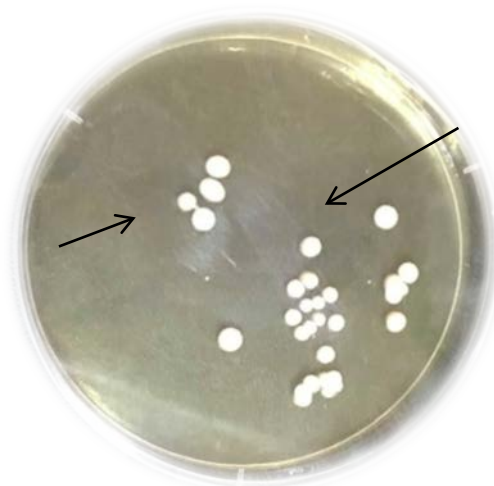


Figure 2. *Candida albicans* cultured on sabouraud dextrose agar.

Microscopic Characteristics :

Fungal isolation gave a positive result with Gram-stine where the oval cells appeared to Spherical or oval to protracted or cylindrical shape. The appearance of cells is colored in blue due to the retention of the peptidoglycan layer in the cell wall of the cell [12] Figure 3.

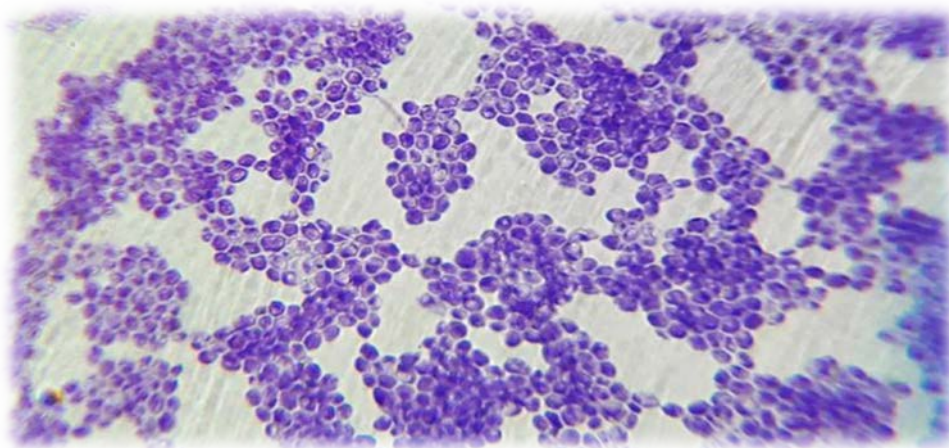


Figure 3. *Candida bicans* under 40x magnification

Germ Tube Formation :

The test results showed that all isolates of the genus *Candida albicans* formed the germ tube at 37 ° C incubation for 2-3 hours in the human serum. However, all other species *C. tropicalis*, *C. parapsilosis*, *C. krusei* were not under the same conditions. The germ tube plays an important role in the permeability of the layer of epithelial cells lining the body and tissues and reaching the bloodstream. In addition, it is believed to be necessary to feed the yeast [13] Figure 4.

After all the previous procedures conducted on the sample proved that the sample belongs to the fungus *Candida albicans*, where we study in this research the effect of extract water and alcohol extract of frankincense on fungi previously diagnosed.



Figure 4. Germ tube of candida

Procedure :**Preparation of watery extracts :**

50 grams of frankincense were dissolved in 250 ml of distilled water and stirred the solution and then left for 24 hours for the purpose of soaking. Then filtered by filter papers with a diameter of 0.6 mm and the solution was poured in a dish and leave until drying after that the samples were preserved after drying in glass bottles with tight cover in moisture-free conditions for use [14].

Preparation of alcoholic extracts :

Prepare the alcohol extract by mixing 50 g of frankincense with 250 ml of ethanol at a concentration of 96% and then follow the same steps after that the product was drying and saved in sterilized bottles until use [15].

Preparation of different concentrations of frankincense extract :

Taking 0.2, 0.4 and 0.8 g of frankincense extract to dissolved in 2 ml of distilled water (for watery extract) and 2 ml of Dimethyl Sulfoxide (for alcoholic extract) to prepare concentrations (100, 200, 400) and their concentrations sterilized by Pasteurization at 62 ° C for 15 min.

Test the effect of extracts on fungi :

A sample of *Candida albicans* was applied to the culture medium (sabouraud dextrose agar) for the purpose of testing the effect of the water and alcohol extracts of the frankincense on the fungus. After The published the sample on the culture medium, incubated the dishes at 37 ° C and for half an hour in the incubator for the purpose of drinking. After that taken a small disks of filter paper diameter 6 mm that was sterilized by autoclave and immersing them in different concentrations of the frankincense extracts (100, 200 and 400). The discs were fixed by sterile forceps on the surface of the fertilized dishes, and the were incubated at 37 ° C for 24 hours. After the incubation, the diameters were observed and measured around the disks saturated with frankincense extract.

RESULTS AND DISCUSSION

According to the results in Table 1 and figure 5 shows the preliminary test of the effect of watery and alcoholic extracts on the fungi studied in the concentration of (200). However, the effect of the alcoholic extract was better compared to the watery extract, of frankincense composition, where several studies confirm the preference of alcohol solvent in the extraction of active substances from medicinal plants [16].

Table 1. The first effect of the extracts

Candida albicans	Fungi	
	Extract	
8.5mm	Watery extract (200) conc.	
15mm	Alcohol extract (200) conc.	



Figure 5. show compare between effective f watery extracts and alcohol extracts at 200 concentrations on the candida.

And to indicate the effect of the Frankincense on the candida, the increased and decreased in the initial focused concentration of a Frankincense substance (200) on its impact capacity has been increased concentration to (400) as the highest concentration and use concentration (100) As a minimum concentration [6, 9].

Table 2 and figure 6 shows the diameters resulting from the use of these concentrations on the studied fungi. It is note in the case of alcoholic extract that the increased or decreased of concentration led to the increase and decrease of inhibitory activity in a clear way and the fungus *Candida albicans* has lost its sensitivity completely in concentration higher than the extract of alcohol, which confirms this frankincense extract contain of the active substances, While in the case of the watery extract, the increased concentration was not accompanied by a significant increase in inhibitory activity, which indicates that water is not the appropriate solvent to extract the active substance found in the composition of frankincense. The increase in inhibitory activity with increased concentration leads to the possibility of using higher concentrations of frankincense extract, especially as some studies confirm that there is no toxicity of this extract when used in laboratory animal experiments. The chemical composition of frankincense is generally composed of volatile oils, glutamic acids and gum, but it is believed that the effectiveness of this substance is due to the presence of Boswlic acid, which acts as an anti-inflammatory in addition to being anti-cancer and allergies and others [17].

Table 2. Extracts for frankincense (diameter inhibition in millimeters)

Candida albicans	Fungi	
	Extract	
7	(100) conc.	Water
9.5	(400) conc.	Extract
12	(100) conc.	Alcohol
19.5	(400) conc.	Extract

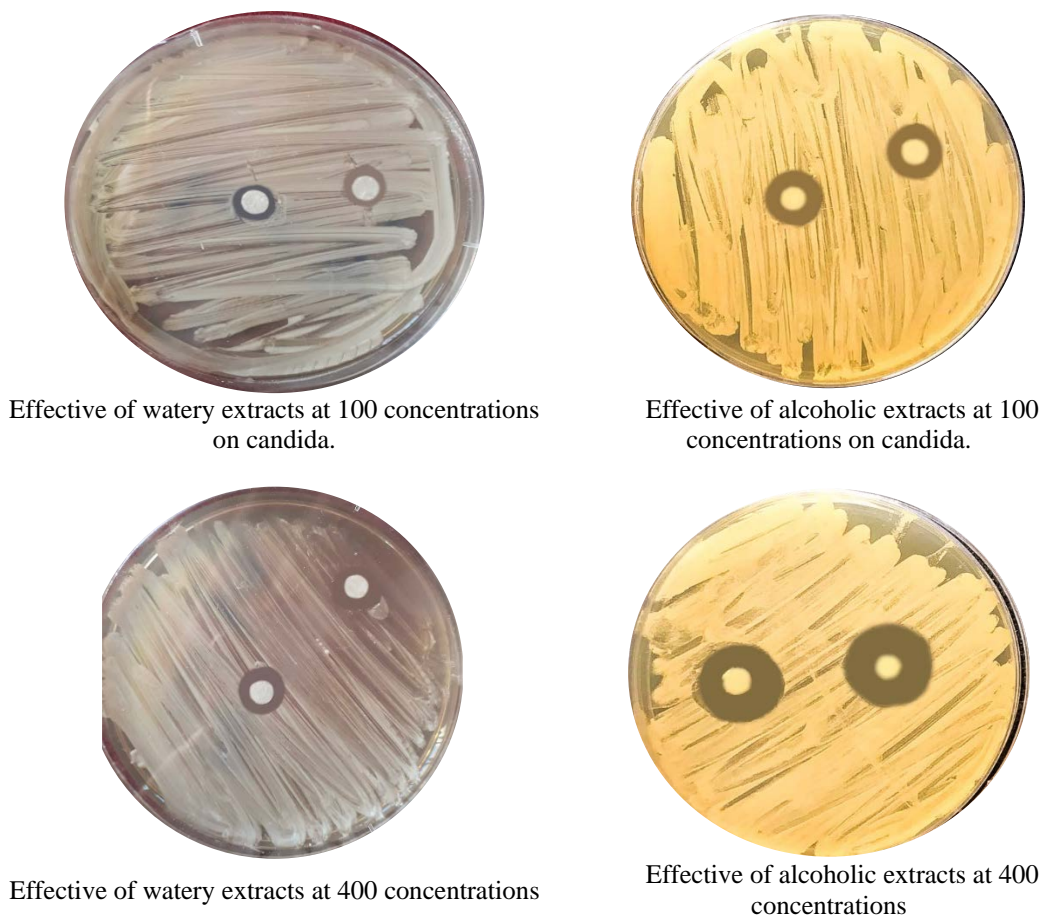


Figure 6. show compare between effective of watery extracts and alcohol extracts different concentration on the candida

Figure 6 view of the effect of various concentrations of watery and alcoholic extracts of frankincense, it has been shown to be competitive with several antibodies affecting *Candida albicans*. In particular, the alcohol extract has a significant effect on fungus figure 7 [18, 19]. *Candida* is causes a serious disease in humans and obtaining a natural antibiotic is very important Especially that the effect of the extract of frankincense exceeds several of the industrial antibiotics, which opens the door to other studies on this or other fungi in order to ensure accurate and appropriate concentration through the study of inhibitory and therapeutic activities in vivo using laboratory animals, as well as conducting analytical studies to know the active component in frankincense towards organisms and perhaps the use of other solvents to extract the active substance and thus fix the therapeutic capacity of these The substance is accurate and therefore can be used as a suitable treatment for sensitive organisms [20, 21].

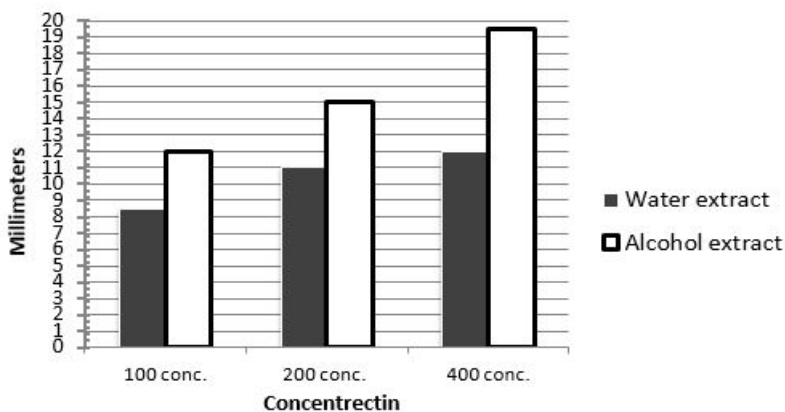


Figure 7. Deferent between effective of alcoholic and watery of frankincense extract on candida albicans (Diameter inhibition in millimetres).

CONCLUSION

Extract of water and the alcohol extract of The frankincense material can be used as an antifungal agent of the candida albicans. It effect the mouth in children and cause the so-called oral cough. The alcohol extract was more effective than the water extract at the same concentrations

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