Antifungal activity of some medicinal plant extracts against some fungal isolates

Actividad antifúngica de algunos extractos de plantas medicinales contra algunos aislados de hongos.

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SCIENTIFIC RESEARCH

How to cite this paper:
Abdullah B A., Salih D T., and Saadullah A. A M., Antifungal activity of some medicinal plant extracts against some fungal isolates. Innovaciencia. 2019; 7 (1): 1-6.
http://dx.doi.org/10.15649/2346075X.506

Reception date:
Received: 25 January 2019
Accepted: 28 May 2019
Published: 25 October 2019

Keywords:
Plant extracts; fungal isolates; Antifungal activity.

ABSTRACT

Introduction: Fungi live everywhere in the environment, most of them are not dangerous, but some types of fungi can be harmful to human health. The medicinal plants contain many antimicrobial components that make them recently used as powerful drugs. The aim of the present investigation was to examine the antifungal potential and minimum inhibitory concentration (MIC) of three plant extract: *Aloe vera* gel, cinnamon (*Cinnamomum zeylanicum*) and turmeric (*Curcuma longa*) against three fungal species*: Aspergillus niger*, *Candida albicans* and *Fusarium oxysporum*. **Materials and Methods**: The plant materials were extracted using solvents DMSO and ethanol and then were tested against the selected fungal isolates using well diffusion method. **Results and Discussion**: Antifungal activity of *Aloe vera* against *Aspergillus niger* showed MIC value of 25% whereas for *Candida albicans* and *Fusarium oxysporum* the MIC obtained was 100%. Both cinnamon and turmeric showed maximum potency against *Aspergillus niger*, *Candida albicans* and *Fusarium oxysporum* at highest MIC value of 100 %. The degree of inhibition increased correspondingly with increasing concentrations of the plant extracts. **Conclusions**: the tested plant extracts have an antifungal activity and could be used as alternative drugs.

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INTRODUCCTION

The use of medicinal plants recently increased significantly and it's shown helpful for primary health care all over the world. Many plants contain antifungal, antibacterial, antiviral and anti-inflammation properties because it contains different kinds of secondary metabolites. Plant secondary metabolites are of low-molecular weight compounds such as phenolic compounds alkaloids, terpenoids, tannins, glycosides, organic acids, resins (including resin acids, resin alcohols and hydrocarbon resins), sugars (including starches, inulin, gums and phlegmatic, amino acids, proteins and enzymes, plant pigments (including chlorophyll, carotenoids, flavonoids, beet red bases and quinones) volatile oils and waxes, and inorganic ingredients (trace elements) [1, 2].

Plant extractions application and multiuse in medical setting has drawn many interests. Microbiologists have used the properties found in phytochemicals as potential antimicrobial drugs clinically. This is especially essential in understanding the alteration of traditional allopathic medical system. Many types of medicinal plants represent a rich source of different kinds of both antifungal and antibacterial agents. Different countries used different kinds of different medicinal plans as powerful drugs. Many parts of medicinal plants are extracting and used as raw drugs because they contain different medicinal properties. These parts include twigs, flowers, fruits, stem, roots and other modified organ of plants [3, 4].

The pathogenic fungi are usually plant, animal and human pathogens and are common in the environment .Fungi live either outdoors on plants and in soil or on many indoor surfaces as well as found on human skin and hair. Most fungi are not dangerous, but some types of fungi can be harmful to human health [5, 6].

The most common fungal species causing Aspergillosis is *Aspergillus fumigatus*, however, other species also can cause the disease such as *Aspergillus niger, Aspergillus flavus* and *Aspergillus terreus* [7]. *Fusarium* is one of the main opportunistic fungi; its toxicity is known by Fusariotoxicosis caused by *Fusarium oxysporum* in many animals. Candidiasis is an opportunistic infectious disease caused by the genus *Candida*, which includes different species. The most common species of the *Candida* is *C. albicans*. Due to the widespread and indiscriminate use of antimicrobial drugs, many microorganisms have developed resistance to specific types of these antimicrobial drugs and these strains are particularly obvious in the hospital environment, this problem also creates massive clinical problems in the treatment of infectious diseases [8, 9].

Repeated consumption of antifungal drugs leads to the development of more resistant fungi and increased damages of great amount of disease spread with side effects. Therefore, it is worthwhile to look for an alternative treatment such as extracting active compounds from many plant species that are used in herbal medicine [10].

Most researches were dedicated to discover and study new natural sources that can suppress pathogenic fungi and replace chemical use of the antifungal agents [11].
The objective of the research was to study the antifungal activity and minimum inhibitory concentration of three plant extract *Aloe vera* gel, cinnamon (*Cinnamomum zeylanicum*) and turmeric (*Curcuma longa*) against three fungal isolates *Aspergillus niger , Fusarium oxysporum* and *Candida albicans.*

MATERIALS AND METHOD

1. Plant materials

Mature leaves of Aloe (*Aloe vera*), turmeric rhizome (*Curcuma longa*) (Zingiberaceae family) and bark of cinnamon (*Cinnamomum zeylanicum*) were obtained from Duhok farms. These plants were botanically authenticated at the College of Agriculture, University of Duhok.

2. Preparation of gel extract from *Aloe vera.*

Mature, healthy and fresh leaves of *Aloe vera* were washed in the running tap water for 5 min and rinsed with sterile distilled water, then dissected longitudinally and the colourless parenchymatous tissue (aloe

gel) was scraped out using a sterile knife without the fibres. The gel was ground with DMSO using the mortar and pestle. The extracts were filtered using Whatman No. 1 filter paper and the filtrate was centrifuged at 5000 RPM for 5 min. The supernatant was collected and stored in refrigerator at 4°C [12]. Different concentration (25%, 50% and 100%) of *A. vera* gel extract was prepared and subjected to antimicrobial studies.

3-Preparation of Ethanolic Extract from cinnamon and turmeric

Sample Preparation Firstly, bark of cinnamon and Rhizomes of turmeric were washed and cut into small pieces. Bark of cinnamon and Rhizomes of turmeric were dried using drying cabinet under 40°C for 3-4 days. The dried plant material was grinded into powder form. 300 g of powdered Cinnamon bark and Rhizomes of turmeric were homogenized using 96% ethanol in a food processor, and then stirred magnetically overnight. The mixture filtrated and the filtrate was evaporated under vacuum to obtain ethanolic extract [13]. Different concentration (25% - 100%) of cinnamon and turmeric extract was prepared and subjected to antimicrobial studies.

4-Fungal strains used

The fungal species used in the present study were *Candida albicans, Fusarium oxysporum* and *Aspergillus niger* which obtained from mycology research lab in the college of science, University of Duhok and previously isolated and identified. Two isolates from each fungal species were tested in this investigation.

5. The antifungal sensitivity test.

The antifungal sensitivity of prepared plant materials were tested against the isolates of three fungal species, each isolate was subcultured before testing in a Sabouraud's Dextrose broth and incubated at 25°C for 24-48 hrs. Potato Dextrose Agar (PDA) medium was prepared and inoculated with 0.1 ml of each of fungal isolates. The sensitivity test was performed using well diffusion method, for each plate a well of 6 mm in diameter was cut out of the agar using cork borer and filled with 0.1 ml of the desired plant extract concentration and incubated at 25°C for 24-48 hrs. [14]. The minimum inhibitory concentration was estimated as the lowest concentration of the plant extract that will inhibit the visible growth of a fungus after the incubation period.

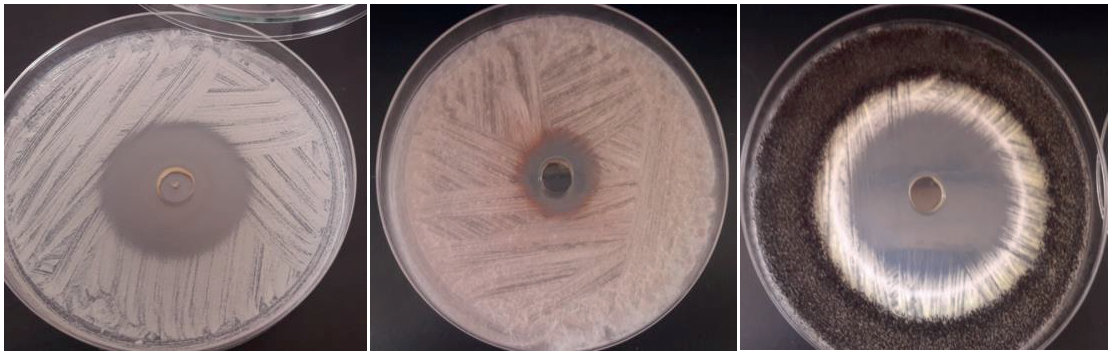
RESULTS

The present study revealed the effectiveness of three plant extracts: *Aloe vera,* turmeric and cinnamon toward selected fungal isolates *Aspergillus niger, Fusarium oxysporum* and *Candida albicans*. Inhibitory effectiveness is varied depending on the type of plant extract and fungal isolate and in general had a high rate of diameters inhibition. DMSO gel extracts of *A. vera* were tested for the antifungal activity against the fungal species and the results are illustrated in the figure 1 and table 1.

The *Aloe vera* gel at concentration of 25% showed high suppression in the growth of *Aspergillus niger* while both *Fusarium oxysporum* and *Candida albicans* inhibited at the concentration of 100%. The result of this study clearly reflect that *Aloe vera* has inherent ability to induce toxic effect on mycelial growth and proliferation of these fungi also *Aloe vera* gel contain saponin which is an soapy substance that has the antimicrobial properties against viruses, bacteria, filamentous fungi and yeasts [15]. The results are in agreement with many findings [16, 17].

Table (1). MIC of *Aloe Vera* against fungal isolates.

No.	Fungal isolates	*Aloe Vera* MIC (%)
1	*Aspergillus niger* (2)	25
2	*Fusarium oxysporum* (2)	100
3	*Candida albicans*(2)	100



(A) (B) (C)

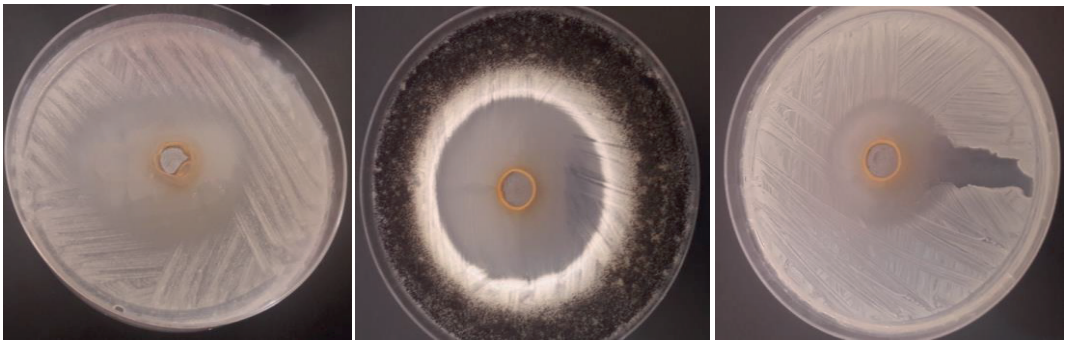
Figure 1. The inhibition zones by *Aloe Vera* with concentration 100 % on different isolates: A. *Candida albicans*. B. *Fusarium oxysporum* and C. *Aspergillus niger*.

Antifungal activity of cinnamon against *Candida albicans, Aspergillus niger* and *Fusarium oxysporum* is given in table 2 and figure 2. Cinnamon showed complete inhibition against three tested fungi at the concentration of 100% .The good potential of cinnamon as antifungal agent was reported previously by many studies [18-21].

Antimicrobial activity of cinnamon has been reported to inhibit the growth of yeasts, molds, and bacteria [22]. The main component of cinnamon is cinnamaldehyde which contains aldehyde group and conjugated double bond outside the ring. Cinnamaldehyde is a powerful antifungal agent effect on fungal cell wall component (1, 3)- glucan and chitin synthesis in molds and yeasts [23, 24].

Table 2. MIC of Cinnamon *(Cinnamomum zeylanicum)* against fungal isolates.

No.	Fungal isolates	Cinnamon *(Cinnamomum zeylanicum)* MIC (%)
1	*Aspergillus niger* (2)	100
2	*Fusarium oxysporum* (2)	100
3	*Candida albicans*(2)	100



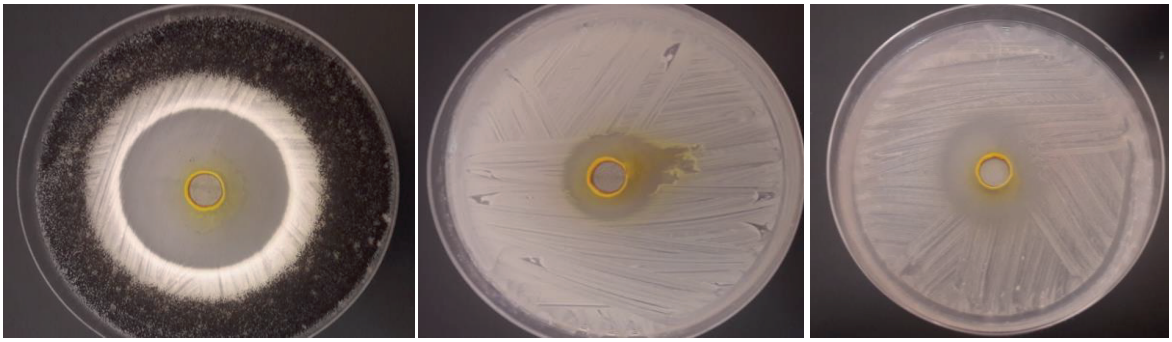
(A) (B) (C)

Figure 2. The inhibition zones by Cinnamon *(Cinnamomum zeylanicum)* at concentration 100 % on different isolates: A. *Fusarium oxysporum* B. *Aspergillus niger* and C. *Candida albicans.*

In the present investigation, in vitro antifungal activity of the DMSO extracts of turmeric was quantitatively evaluated according to the zone of inhibition. The concentration of 100 % DMSO extracts of turmeric studied in the present investigation exhibited inhibitory effect against the three selected human fungal pathogens (Table and figure 1). Because of the traditional extensive use of turmeric in food products, many researchers have been done to study the turmeric to controlling fungal causing spoilage and fungal pathogens [25].

Table 3. MIC of Turmeric *(Curcuma longa)* against fungal isolates.

No.	Fungal isolates	Turmeric *(Curcuma longa)* MIC (%)
1	*Aspergillus niger* (2)	100
2	*Fusarium oxysporum* (2)	100
3	*Candida albicans*(2)	100



A B C

Figure 3. The inhibition zones by Turmeric *(Curcuma longa)* with concentration 100 % on different isolates: A. *Aspergillus niger*, B. *Candida albicans*, C. *Fusarium oxysporum*.

CONCLUSIONS

We conclude from the current study the antifungal activity of three tested plant extracts showed prominent antifungal activity against the fungal isolates *Aspergillus niger, Candida albicans* and *Fusarium oxysporum* and contain more active compounds allowing recommended therapeutic alternatives to antifungal chemical agents in addition the plants extracts are source of cheap antifungal.

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