

EFFECT OF ERUCA SATIVA (ROCKET SALAD) EXTRACT ON THE GROWTH OF CANDIDA ALBICANS IN VITRO

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Abstract

Plant extracts have been used for a long time as good alternatives to antibiotics in the inhibition of many microorganisms as they are not polluted and does not develop any kind of resistance, and watercress plant is one of those common plants in Iraq, used for medical and industrial purposes and since ancient times, this study came to test the effect of *Eruca sativa* (Rocket) extract on the growth of *Candida albicans* yeast as one of the most important fungi that cause many diseases in humans and animals. Aqueous extract of *Eruca sativa* prepared by boiling of the plant leaves in the water and using the filtered extract directly to checking the effect of this extract on the pathogenic isolate of *Candida albicans* by agar diffusion method and also testing the effects of the prepared extract on the *Candida albicans* structure using scanning electron microscope (SEM). The results of this study proved that the *Eruca sativa* aquatic extract had a inhibitory effect on the growth of yeast on the laboratory media.

Keywords: Aqueous extract, Candida albicans, Eruca sativa.

Introduction

Rocket plant belongs to Angiosperms plants. Dicotyledons, where hails from Brassicales order, (Cruciferae) Brassicaceae family, old scientific name Brassica eruca L. and modern name Eruca sativa Mill. (Blamey and Wilson, 1989). Eruca sativa Mill. It is believed that its native place in Central Asia and Eastern Europe, and Rocket salad leaves good nutritional value is evident when comparing their nutritional content with some fresh leafy greens, especially in the content of calories, total oil, carbohydrates and some nutrients, use fresh green leaves with distinctive taste salads in many countries of the world. so the plant is called Rocket salad. The leaves may be dried to be used as a food spice or condiment. Fresh or dried leaves may be eaten for their medicinal effect in increasing diuresis (Mahran et al., 1991). This plant also as activation for blood circulation, Anti-inflammatory skin (Mohamedien, 1994; Hila et al., 2009). Antiscorbutic, anti-stomach pain, Sexual stimulant, antifungal foot (Bhandari and Chandel, 1996) Laxative, Digest, Tonic, Emollient, Astringent, Rubefacient and antiperspirant (Ambasta, 1986). Rocket salad oil also has medical efficacy that Produced in the laboratory. These include improved liver function, increased sperm fertility, improved progesterone, estrogen and sexual glands, weightloss programs (Merza et al., 2000), menstrual depurative, antidiabetic, antihyperlipidemic, antibacterial, antifungal and a catalyst for vomiting (Khare, 2007).

Candida albicans is a medically importance opportunistic fungus can cause many diseases in humans and animals (Correia *et al.*, 2004) and considered as one of the three leading causes of hospital infection in the United States of America (Wisplinghoff *et al.*, 2004) in addition to the economic losses due to spreading of this microorganisms in the hospitals and also in the food industry (Wilson *et al.*, 2002).

The effects of some plant extracts on the inhibition of the growth of some fungi (Abu-shanab *et al.*, 2005; Peter *et al.*, 2009; Gulfraz *et al.*, 2011) have been studied. However, few of these studies focused on the effect of Rocket plant extracts on the *Candida albicans* growth and structure, this study aimed to investigate the effect of *Eruca sativa* extract on the growth of *Candida albicans* in vitro.

Material and Methods

Plant extraction preparation

The method adopted by (Metsulpa *et al.*, 2001) in the preparation of aqueous extract of *Eruca sativa* leaves, where taken (10g) of fresh soft leaf powder and placed in a glass flask (500ml) contains (200ml) distilled water, mixed the plant material with a magnetic mixer for (15min) and then leave the solution for (24hours) to precipitate the plant parts, and after (3hours) the solution was filtered and the sediment was neglected and the filtrate was taken and a filtrate was obtained and used later as Stock solution.

Yeast preparation

The study was carried on a pathogenic isolate of *Candida albicans* obtained from clinical case from previous study (Samaka et al., 2018). The sensitivity of the Candida albicans to the Eruca sativa aqueous extract was tested on the Candida CHROMagar medium (CHROMagar microbiology, France) according to the CLSI- M44-A2 guideline by spreading of yeast suspension equivalent to the McFarland No. 0.5 tube $(1-5 \times 10^6 \text{ yeast cells/ml})$ on the medium using a cotton swab (CLSI, 2009), about 100 microliters of the Eruca sativa aquatic extract in a one well on the medium while 100 microliter of distilled water and 100 microliter of garlic (Allium sativa) extract (commercially prepared extract from EMAD company, Iraq) were used as a negative and positive control respectively, where, the inhibitory effect of the garlic extract is known on the Candida albicans (Al-Saimary, 1999). Plates were incubated at 25°C for 48 h. and growth was compared to that in the control well. The experiment was repeated triple to confirm the results. After 24h incubation, the inhibitory effect was observed and the diameter of the inhibition zone was measured on the culture medium compared to the positive and negative control wells.

Effect of *Eruca sativa* extract on the *Candida albicans* structure:

Two group of plates were prepared for testing the effect of *Eruca sativa* plant extract on the *Candida albicans*. Group I (GI) represent a yeast cells grow on the Candida CHROMagar medium (Candida CHROMagar microbiology, France) while the second group (GII) represent a yeast cells exposed to the aquatic extract of *Eruca sativa* plant. All plates were incubated at 25 °C for 24 hours, yeasts from the two groups were collected and examined under a scanning electron microscope (SEM) at the Faculty of Science at the University of Kufa, to detect the structural changes in the study isolate regarding to exposure to the *Eruca sativa* extract.

Results and Discussion

The results showed that the aquatic *Eruca sativa* extract has an inhibitory effect on the growth of yeast *Candida albicans in vitro* (figure 1), and this is consistent with (Amna *et al.*, 2014) due to the presence of a number of compounds such as (flavonoids, alkaloids, tannins, phenols, saponins and ascorbic acid) in Eruca extract, which has a inhibitory effect on other organisms such as some fungi and bacteria (Gulfraz *et al.*, 2011).

First group (control group)



Fig. 1: Shows inhibitory effect of *Eruca sativa* Plant extract on growth of *Candida albicans* yeast on CHROMagar, where : A) The test hole, contains Rocket salad plant extract. B) a positive control hole, contains garlic plant extract. C) a negative control hole, contains distilled water.

Electron microscopy parameter

The scanning electron microscopy images, showed that there were clear changes within the cell wall and cell structure of *Candida albicans* yeasts that led to the damage of the yeast cells compared to the control group (figure 2).

Second group (treatment group)



Fig. 2 : Showing the effect of *Eurca sativa* extract in the *Candida albicans* grown on the CHROMagar medium, Where: The first group, represents the control group. The second group, represents cells exposed to *Eurca sativa* extract.

Conclusion

- The presence of high concentrations of phenolic compounds in *Eruca sativa* extract (Pasinia *et al.*, 2012) led to the direct effect on the cell wall of the yeast under study. Previous studies have also shown that phenolic compounds have the potential to cause holes in the cell wall of yeasts and damage them (Martins *et al.*, 2015).
- So, we can conclude that the effect of *Eruca sativa* extract on the damage to the yeast wall of *Candida albicans* is the main factor in inhibiting the growth of these yeasts on the media.

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