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RESEARCH ARTICLE

CUCURBITA MOSCHATA: A PLANT WITH ANTIBACTERIAL PROPERTIES

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ABSTRACT

Cucurbita moschata (CM) is an indigenous plant in Iran, which the plant has been applied as an antihypertensive, anticancer, antiparasitic, antiviral, antifungal, antioxidant, and antiflammatory agent in Iran. In comparison to many other pharmaceutical-industrial plants, there is a very little data about antibacterial activities of CM oil collected from Iran. Hence, the aim of the current study was evaluation of antibacterial activities of the oil on *Escherichia coli O157:H7* (ATCC No. 25922) (EC) in west of Iran (In Kermanshah). The antibacterial effects of CM oil was assessed by macro-dilution method in Mueller-Hinton broth medium, agar disk and well diffusion methods. The results demonstrated that the oil of CM have prevented the growth of EC and killed it. Also, by incrementing the concentration of the CM oil, the inhibition zones incremented. We believe that the article provide support to the antibacterial properties of the oil. In fact, this work indicate that the oil of CM can be beneficial as medicinal composition. Fractionation of active molecules will be the future work to peruse.

INTRODUCTION

From past to now, People used from ethnomedicinal plants for the inhibition and cure of varied diseases such as bacterial infections[1-4]. Because of their safety and low cost as well as their effect on a wide variety of bacteria, ethnomedicinal plants may have the potency to treat bacterial resistance to various types of antibiotics[5-7]. Gram-positive (such as *Staphylococcus aureus* and *Bacillus subtillis*) Gram-negative (*Pseudomonas aeruginosa* and *Escherichia coli*) bacteria are very sensitive to plants[8-11]. Plant derived products have a main versatility of phytochemicals compounds[12]. The results indicated that these mechanisms of antibacterial effects of plants vary greatly depending on the components of the plant[13, 14]. Oils are potential sources of new antibacterial compounds, especially on bacterial pathogens[15]. *In vitro* studies in the work demonstrated that the oils prevented bacterial growth but their effectiveness varied[16]. The antibacterial properties of many oils has been previously reviewed and classified as strong, medium or weak[17].

Cucurbita moschata (CM) is part of the *Cucurbitales* order, *Cucurbitaceae* family and *Cucurbita* genuse that has long been applied in Asia for medicinal goals[18]. CM seeds has many nutrients including CM polysaccharides, essential fatty acid, carotenoids, mineral, active proteins, and essential amino acids. Due to the compounds mentioned CM seeds has a high nutritional value[19]. From the past, demonstrated that seeds of CM had medicinal properties for their biological effects such as antimicrobial activities. The antibacterial properties of CM seeds has many applications such as alternative medicine and innate therapies[20].

The aim of this study was to screen the in vitro antibacterial properties of the plant oil against *Escherichia coli O157:H7* (ATCC No. 25922) (EC). Exploring the scientific justifications behind its traditional use could defend to detect the probability to develop further new biotechnological benefits.

MATERIALS AND METHODS

Source of microorganisms

Escherichia coli O157:H7 (ATCC No. 25922) (EC) was purveyed by The Iranian Research Organization of Science and Technology. The strain was activated on Tryptic Soy broth kept at 37°C for 18 h. 60 µl broth was transferred to Nutrient agar and incubated at 37°C for another 24 h; cell concentration was adjusted to get a final concentration of 10^8 cfu/ml in Muller Hinton broth.

Culture media

Mueller-Hinton Agar growth media was aggregated according to manufacturer's instruction (Oxoid, UK), autoclaved and dispensed at 20 ml per plate in 12 x 12cm Petri dishes.

Essential oil extraction

The crushed CM seeds were dried in an oven at 105°C to clear any moisture presence at several interval of time. A 10 g of dried CM seeds was placed onto a thimble and the thimble was put into the sohxlet extractor. N-hexane solvent was infused into three-neck- round bottom flask that is joined with the extractor and flask along with the condenser on the top to eschew any solvent losses. The lump assembly was then set on the temperature controller heater to furnish the required temperature. The temperature was measured by a thermometer that was inserted in one of the necks of the round bottom flask. After distinctive interval of the time the tryout was stopped and the trapped oil in the solvent was separated. The mixture of solvent and oil was separated using rotary evaporator under vacuum at temperature of 65°C.

Then, oil obtained after evaporation was weighed.

Evaluation of antimicrobial activities

Agar disk and agar well diffusion tests were done to calculate Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) of CM seeds oil to prevent EC. The solution of CM seeds produced in 1ml from which six fold serial dilutions (v/v) of 60 µl were placed on each disk and well. Dimethyl sulfoxide (DMSO) was applied as negative control and Kanamycin as a positive control. After 24 hours incubation diameters of growth inhibition zones around disks or wells were measured. For specification of MIC, the macrobroth dilution method was used[21].

RESULTS

About CM, the widest zone was seen in 0.125 g/ml concentration (The value of growth inhibition zone was 15 mm in this dilution).

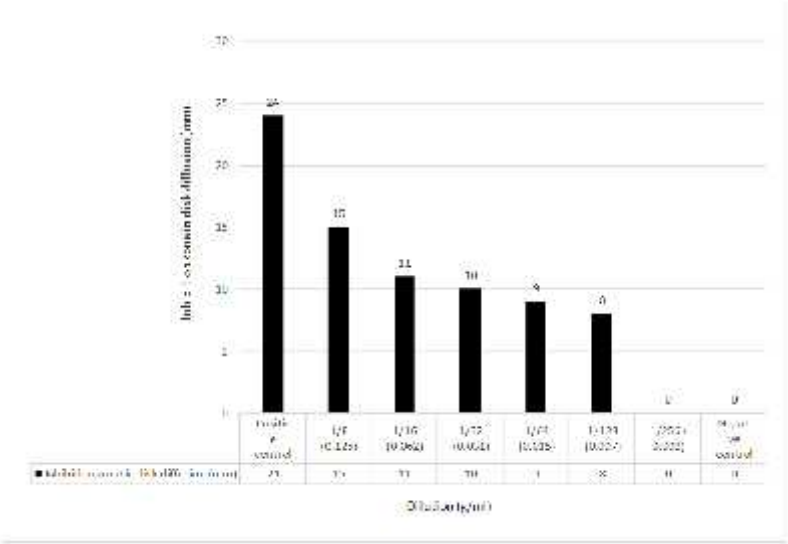

Figure 1 The diameters of growth inhibition zones in agar disk diffusion test in different dilutions of CM.

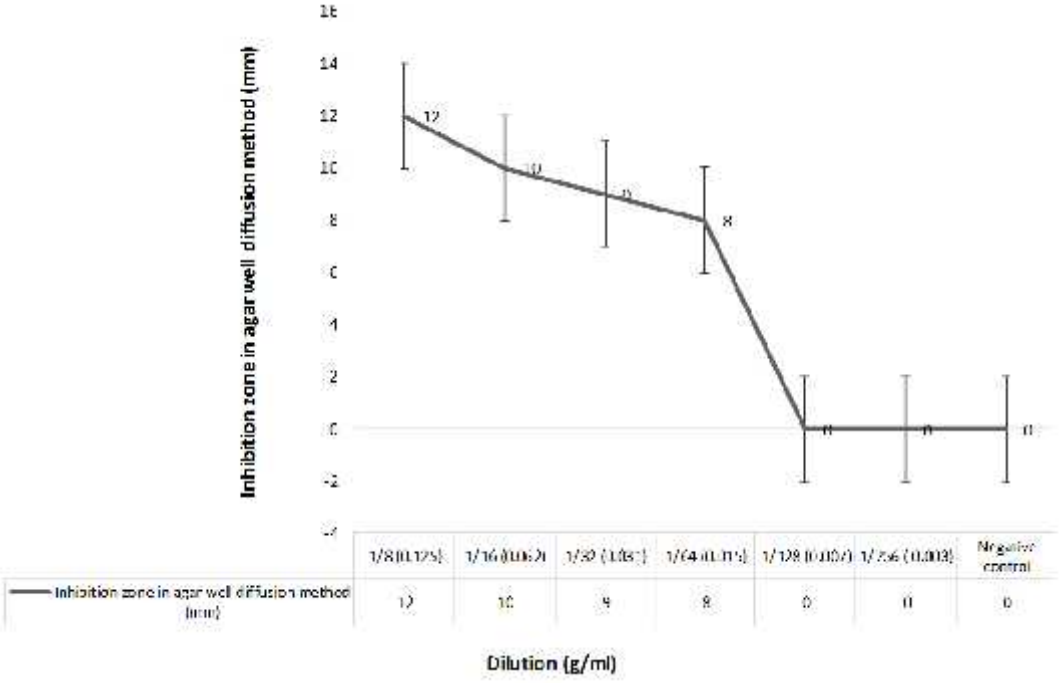

Figure 2 The diameters of growth inhibition zones in agar well diffusion test in different dilutions of CM.

No inhibition zone was observed due DMSO. Growth inhibition zones due to different dilutions are listed in figure 1.

In regard to CM, the widest zone was seen in 0.125 g/ml concentration (The diameter of growth inhibition zone was 12 mm in this dilution). There was no inhibition zone in EC due to 0.003 and 0.007 g/ml concentrations. No inhibition zone was observed due to DMSO. The data are discoverable in figure 2.

In the examined bacterium, MIC and MBC values were the same and equal to 0.062 g/ml concentration.

DISCUSSION

Most bacteria has become persistent to ordinary antibiotics and plants and their derivatives are being screened as alternatives[22-25]. Plant have been screened for their possible uses as better orders for the therapy of different bacterial infections[26, 27]. In Iranian ethnomedicine, plant in the different form are applied by the population for the cure of diseases including bacterial illness[28-30]. In Iran, a plant with the scientific name of CM has traditional medical usage such as avails in the remedy of bacterial infections.

The antibacterial results demonstrated that the CM seeds inhibited the bacterium and the effects were dependent upon concentration. In agar well diffusion test, the widest zone was seen in 0.125 g/ml concentration (12 mm) and no inhibitory activity of seeds of CM in 0.003, and 0.007 g/ml concentrations. In agar disk diffusion test, the widest inhibition zone was seen in 0.125 g/ml concentration (The value of growth inhibition zone was 15 mm in this dilution, but the value of growth inhibition zone of Kanamycin against EC was 24 mm) and no inhibitory effects of DMSO against the EC. Also, the results indicated that CM seeds with 0.062 g/ml concentration has prevented from the growth EC, also in this concentration has killed. There are correspondences between these results and the resembling studies. In these similar papers, indicated that CM seeds have strong antibacterial properties[20, 31.] Our results assert the use of this plant in traditional medicine and suggest that CM seeds have good antibacterial effects. It can be applied as antibacterial supplement in the developing countries towards the development of recent therapeutic agent. Additional *in vivo* studies and clinical trials would be needed to justify and further assess the potential of the plant as an antibacterial agent in medical purposes.

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