

## Research Paper

# The Antiamoebic Effect of *Oliveria Decumbens* and *Peganum Harmala* Extract on *Acanthamoeba*




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## ABSTRACT

**Background and Aim** *Acanthamoeba* is one of free-living amoebas, which are very abundant in nature. As a free-living amoeba, this parasite has a very high lethality, especially in people with underlying diseases, so researchers are always looking for a way to combat it. Drug plants are a good way to fight *Acanthamoeba* species. In this study, we aimed to investigate the lethal effect of the extract of *Oliveria decumbens* vent and *Peganum harmala* alcoholic extract on *Acanthamoeba*.

**Methods & Materials** In this study using the extract from an extract of *Oliveria decumbens* vent and *Peganum harmala* with concentrations of 1.25, 2.5, 5, 10, and 20 mg/ml to investigate the lethal effect of this extract. The plant was treated with *Acanthamoeba* amoebae after three times (24, 48, and 72) hours.

**Ethical Considerations** This study was approved by the Ethics Committee of the Jahrom University of Medical Sciences (Code: IR.JUMS.REC.1398.029).

**Results** The present research showed that using different concentrations at three times (24, 48, and 72) hours the effect of the extract on trophozoites and cysts of *Acanthamoeba* was shown. The highest lethality is related to the concentration of 20 mg/ml using a combination of both extracts at the time 72 hours and the lowest lethality is related to the concentration of 1.25 mg/ml of *Oliveria decumbens* vent at the time 24 hours.

**Conclusion** Observations indicate that the alcoholic extract of *Oliveria decumbens* vent and *Peganum harmala* had a perfect effect both separately and in a combination of both extracts. These two extracts had a synergistic effect on the lethal effect of *Acanthamoeba* amoeba.

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## Extended Abstract

### Introduction

**F**ree-living amoebas are protozoans that are abundant in nature. [1]. Some species of these amoebas cause serious and sometimes fatal diseases in humans and animals. [2, 3].

The high lethality of this disease has caused researchers to always look for a way to fight free-living amoebas [4]. Various plants have been researched to fight free-living amoebae, each of which has a different range of lethal effects on these protozoa according to the type of plant [5-7]. *Peganum harmala* is an herbaceous plant and a bush whose height reaches up to 50 cm. *Peganum harmala* is found in most places [8]. Recently, there has been an increase in research on plants, such as *Oliveria decumbens* and *Peganum harmala*, whose germicidal effect has been proven; thus, we decided to study the anti-*Acanthamoeba* effects of the alcoholic extract of *Oliveria decumbens* and *Peganum harmala*.

### Materials and Methods

*Oliveria decumbens* and *Peganum harmala* were collected from the southern regions of the country and dried in suitable conditions away from the sun. Then, the extracts were obtained using a percolation device and dried. In the next step, the desired amounts of each of the dried extracts were dissolved in distilled water to obtain different concentrations [9]. *Acanthamoeba* was cultured on non-nutrient agar plates. After almost three weeks, the amoebae were washed, then trophozoites and cysts were counted using trypan blue dye and Nicobar slide. To evaluate the anti-amoebic activity of the extracts in their different concentrations, which were prepared as 1.25, 2.5, 5, 10, and 20 mg/ml, were added separately to the microtubes containing *Acanthamoeba* [10]. The effect of the extracts in different dilutions was investigated at 24, 48 and 72 hours [9]. The collected data were measured using SPSS 21 software.

### Results

The highest lethality was related to the concentration of 20 mg/ml using a combination of both extracts at 72 hours and the lowest lethality was related to the concentration of 1.25 mg/ml of *Oliveria decumbens* at 24 hours. As seen, the results showed the lethal effect of *Oliveria decumbens* and *Peganum harmala* extracts on trophozoites and cysts of *Acanthamoeba*, and the lethal effect of the extracts had a direct relationship with the concentration and duration of the effect, so that as the amount and time increased, the

amount of amoebae decreased and showed a significant difference ( $P < 0.05$ ). Also, the results indicated the greater effect of *Oliveria decumbens* extract and the synergistic effect of these two extracts on each other.

### Discussion

Many studies have been done worldwide to find a suitable method to prevent *Acanthamoeba* infection, and medicinal plants are always considered a suitable method to prevent and treat diseases [5, 11]. Nayeri et al. investigated the effect of artemisinin and alcoholic and aqueous extracts of *Gondwash* (*Artemisia annua*) on *Acanthamoeba* T4 genotype in vitro. They showed that different concentrations of *Artemisia annua* extract have anti-*Acanthamoeba* activity and in the presence of 10 mg per ml of alcoholic extract in the culture medium after 72 hours, 30.51% of trophozoites were alive. Also, in the presence of 10 mg/ml aqueous extract of *Artemisia annua* in the culture medium, after 72 hours, 58.25% of trophozoites were alive, which was consistent with the results of this study and the lethal effect of *Peganum harmala* alcoholic extract. Also, considering the percentage of lethality related to *Peganum harmala*, it can be said that the lethal effect of the *Peganum harmala* alcoholic extract was greater than that of *Artemisia annua* extract [9]. Nayeri et al investigated the aqueous extract of *Peganum harmala* and reported that the aqueous extract of *Peganum harmala* has a dose- and time-dependent anti-amoebic activity on trophozoites and cysts, and in the presence of 10 mg/ml of aqueous extract of *Peganum harmala* in the culture medium after 72 hours, 47.32% of trophozoites and 15.50% of cysts were alive [9, 12]. Despite the obtained results, it can be said that the alcoholic extract of *Oliveria decumbens* has a greater amoebic effect than the extract of the *Peganum harmala* and also these two plants had a synergistic effect on each other.

Therefore, it can be said that *Oliveria decumbens* and *Peganum harmala* have a good killing effect on *Acanthamoeba* amoeba. If they are used together, they increase each other's killing impact.

### Ethical Considerations

#### Compliance with ethical guidelines

This study was approved by the Ethics Committee of the Jahrom University of Medical Sciences (Code: IR.JUMS.REC.1398.029).

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### **Authors' contributions**

All authors equally contributed to preparing this article.

### **Conflicts of interest**

The authors declared no conflict of interest.