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# Use of Essential Oils as Bioherbicides

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**Abstract:** A study was conducted to assess the allelopathic effects of essential oils from Eucalypt, Lawson Cypress, Rosemary and White cedar with the objective of exploring the possibility of their utilization for future weed management. The effects of these four essential oils on the germination of three weed species were examined. The essential oils severely affected the germination percentage of the weed species and showed allelopathic potential. The results demonstrated that germination of the weed species was strongly inhibited by all essential oils, especially Eucalypt oil, when applied at 300 ppm. Results showed that the use of essential oils can be applied for biological control of weeds as pre-emergent weed seed germination inhibitors and, they can decrease the use of chemical herbicides. The possible use of essential oils as herbicides is discussed.

**Key Words:** Allelochemical, essential oils, herbicides, germination inhibitor, weeds.

**Introduction:** The Amaranth, Purslane and Knapweed species are very widespread weeds that decrease the yields of almost all vegetables and fruit orchards in the south of Iran [31]. Potential damage to human health and to the environment from herbicides is regarded as a real problem and this has resulted in an increased interest in alternative strategies leading to the development of biodegradable compounds [11,14,23,31,38]. Inhibition of the growth of plants by other plants in their vicinity has been known for a long time and the chemical interaction between plants, which can cause enhancement or inhibition of growth, has been named allelopathy [25,33]. Evidence for allelopathic interactions in nature by plants containing volatile allelochemicals has been frequently described [1, 8, 12, 13, 24, 27, 30, 31, 32, 36, 37, 43]

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Allelopathic competence of leaves and roots of many plants and trees is well documented in both laboratory and greenhouse experiments. Most of the germination and growth inhibitors produced by perennial angiosperms identified by Rice [33] were phenolic compounds or derivatives of cinnamic acid. Other authors also found coumarins, flavonoids, alkaloids, terpenes, cineoles, citronellal, cyano glycosides, proteins and amino acids among the inhibitory compounds [4, 5, 7, 10, 16, 19, 20, 27, 28, 29, 41, 42].

Muller *et al.*[26] and Friedman *et al.*[21] found that volatile foliage compounds were the active ingredients causing the repression of growth in the vicinity of the *Salvia* and *Artemisia* species. This ecological phenomenon provided a competitive advantage to aromatic plants in their natural environments. Singh *et al* [38] also reported the herbicidal activity of volatile oils from *Eucalyptus citriodora* against *Parthenium hysterophorus*. These reports suggest that allelochemicals could be used for weed control in agriculture.

In the present study we report on the activity of essential oils as germination inhibitors of some important weed species and their possible use as herbicide.

## Experimental

**Extraction of Essentials Oils:** The aerial parts of Eucalypt (*Eucalyptus nicholii*), Rosemary (*Rosmarinus officinalis* L.), Lawson cypress (*Chamaecyparis lowsoniana*) and White cedar (*Thuja occidentalis*) plants were collected in the morning period during July from the landscape of the Shahed University, Tehran, Iran. The aerial parts of investigated plants were air dried at room temperature (less than 25°C) in a shady location for 10-16 days dependent on plant species. The dried herb of all species was subjected to water distillation (hydro distillation) for 3 h using an all glass Clevenger-type apparatus, to extract oil according to the method recommended by the European Pharmacopoeia [18]. The extracted oils were dried over anhydrous sodium sulphate and stored in sealed vials at low temperature before the experiments commenced.

### Bioassay of Inhibition Induced by Essential Oils

**Experiment 1:** The inhibitory effect of the four essential oils was investigated in a preliminary test. Lettuce seeds were germinated on two layers of wet filter paper (Whatman No.3) with 0, 100, 200, 300 and 400 ppm of the four essential oils listed above. Petri dishes containing 50 seeds were incubated in the dark at 27°C. After incubation, the number of germinated seeds were counted in the control for 5 days and compared with the number of seeds from the essential oil treated dishes.

**Experiment 2:** The Seeds of weed species of Amaranth *(Amaranthus retroflexus)*, Purslane (*Portulaca oleracea*) and knapweed *(Acroptilon repens)* were germinated in Petri dishes (9 cm diam.) on two layers of filter paper (Whitman No.3) wetted with 5 ml of distilled water and essential oil. Petri dishes which contained 50 seeds were incubated in the dark at 27°C.

For assessment of the inhibitory effect of essential oils on the seed germination of weeds, concentrations of 0, 100, 200, 300 and 400 ppm were prepared by ether solvent. After incubation, the numbers of germinated seeds were counted for 30 days.

**Statistical Analysis:** The experiments were arranged as a completely randomized design with four replications of each treatment. The significance of differences ($P < 0.05$, 0.01) between treatments was determined by Tukey multiple range tests. All the statistical analyses were performed using SPSS/PC software version 13.

**Results and Discussion:** The allelopathic effects of the essential oils from Eucalypt, Rosemary, Lawson Cypress and White Cedar on the germination percentage of various weed species were determined. The essential oils resulted in decreasing lettuce seed germination as shown in Figure 1.

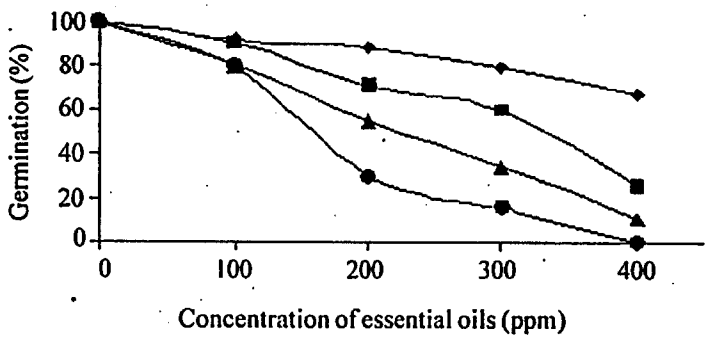


**Fig 1.** The effect of essential oils on the germination of lettuce seeds ●, *Eucalyptus nicholii*; ▲, *Rosmarinus officinalis;* ■, *Chamaecyparis lowsoniana* ◆, *Thuja occidentalis*

The results showed that germination percentage of weeds varied with different concentrations of essential oils. The control treatment showed the maximum germination percentage and the different essential oils exhibited various effects in controlling of germination percent of weed species (Table 1).

The 400 ppm concentration of Rosemary essential oil resulted in a decrease of weed germination percentage and the maximum inhibition effect was observed with Rosemary essential oil on Amaranth seed germination. There were also no significant difference between control and 100 ppm of Rosemary essential oil on the germination of weeds ($P<.05$) (table 1 and Fig 2).

Eucalypt essential oil exhibited more controlling effect than other essential oils and caused severe decrease of three weed species germination. However, the 300 ppm concentration of Eucalypt essential oil showed greatest inhibition effect ($P<.01$). A significant difference ($P<.01$) was not observed between 300 and 400 ppm (Table 1 and Fig 3).

The Lawson Cypress essential oil also showed inhibitory effect on germination of the studied weed species as 300 ppm resulted in significant decrease of weed germination and showed more inhibitory effect on Purslane and Knapweed seeds (Fig 4). Significant difference ($P<.01$) was also observed between 100 ppm and control treatment of Lawson Cypress essential oil (Table 1).

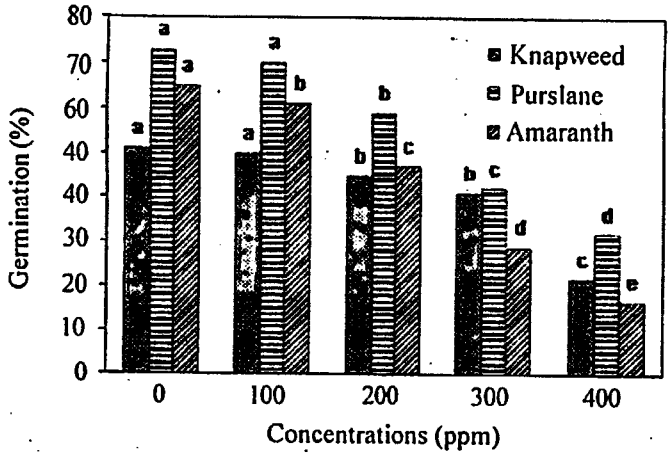


**Fig 2.** Effect of various concentrations of Rosemary essential oil on seed germination of three weed species

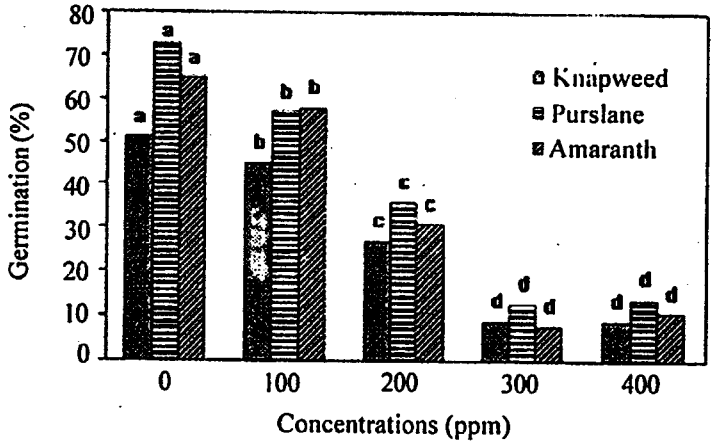


**Fig 3.** Effect of various concentrations of Eucalypt essential oil on seed germination of three weed species

Since germination percentage of weeds at the highest concentration of White cedar essential oil (400 ppm) in comparison with the same concentration of other essential oils was higher, therefore it can be concluded that White cedar essential oil has low effect on seed germination (Table 1). However, the 400 ppm concentration showed significant difference with control and other concentrations. The results also showed that White cedar essential oil has low and similar effect on three weed species (Fig 5).

The essential oils from four plants, *Eucalyptus nicholii, Rosmarinus officinalis, chamaecyparis lowsoniana* and *Thuja occidentalis*, were effective in inhibiting germination at the evaluated concentrations. However, the seeds of some weed species were

much less sensitive to certain essential oils as compared with control and other essential oils. The results of this study therefore showed that there are differences in the sensitivity of the various weed species. There are also differences in herbicidal potency of the essential oils.

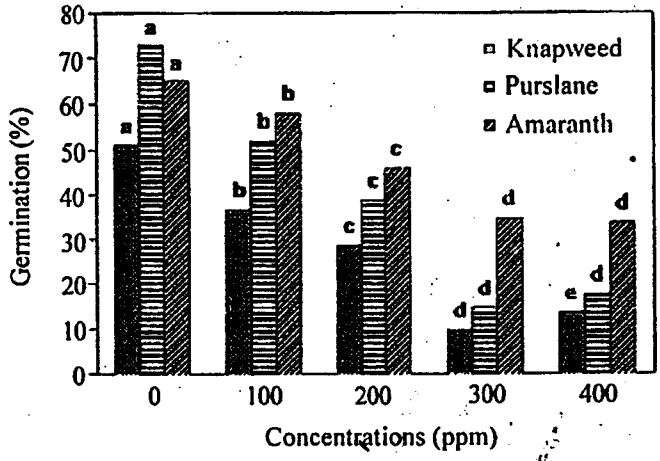


**Fig 4.** Effect of various concentrations of Lawson Cypress essential oil on seed germination of three weed species

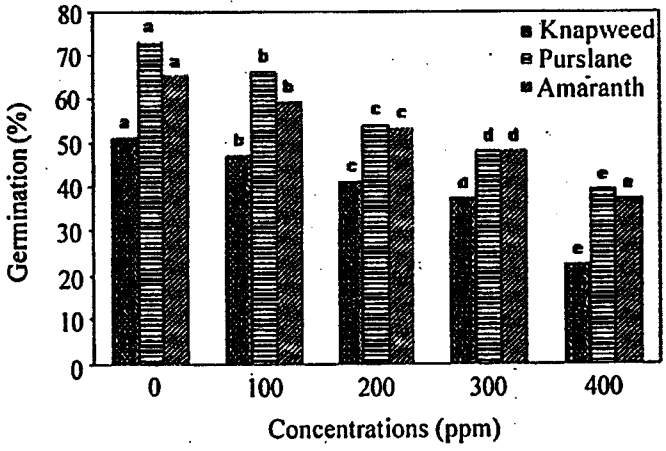


**Fig 5.** Effect of various concentrations of White Cedar essential oil on seed germination of three weed species

Most investigations on allelopathy have characterized the phenomenon, but did not consider their applications in agriculture. In the present study we have examined the first steps towards a possible practical application. However, some studies have indicated that volatile monoterpenes are potent inhibitors of mitosis[6,34]. Vaughn[40] reported that essential

oils from cinnamon (*Cinamommum zeylanicum* Blume) and Red thyme (*Thymus vulgaris* L.) inhibit potato sprout growth by killing meristematic cells. This aspect may need further study.

Previous reports in the literature have shown that the essential oils of the investigated plants in this study contained abundant amounts of monoterpenes and phenolic components, which show inhibitory effects [2,3,9,15,17,22,35,39,45,46,47]. From the present study, it could therefore be concluded that the inhibitory effects of the essential oils on seed germination of weeds could well be attributable to these components.

We suggest that use of Rosemary, Eucalypt, Lawson Cypress and White Cedar essential oils at 400, 300, 300 and 400 ppm concentrations respectively could be applied for control of seed germination of the four weed species in this study. The results of this study also suggested that essential oils could be used for biological control of weeds as pre emergence. Further studies, especially on formulation of essential oils, are still required to apply this technique to agriculture.

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**Table1. The effect of various concentrations of essential oils on the germination of weeds**

| Aromatic plants | Weed species | Essential oil concentration (ppm) | | | | |
|---|---|---|---|---|---|---|
| | | 0 | 100 | 200 | 300 | 400 |
| Rosemary | Knapweed | 51 a | 50 a | 45 b | 41 b | 22 c |
| | Purslane | 73 a | 70 a | 59 b | 42 c | 32 d |
| | Amaranth | 65 a | 61 b | 47 c | 29 d | 17 e |
| Eucalypt | Knapweed | 51 a | 45 b | 27 c | 9 d | 9 d |
| | Purslane | 73 a | 57 b | 36 c | 13 d | 12 d |
| | Amaranth | 65 a | 58 b | 31 c | 10 d | 10 d |
| Lawson cypress | Knapweed | 51 a | 37 b | 29 c | 10 d | 14 e |
| | Purslane | 73 a | 52 b | 39 c | 15 d | 18 d |
| | Amaranth | 65 a | 58 b | 46 c | 35 d | 34 d |
| White cedar | Knapweed | 51 a | 47 b | 41 c | 37 d | 22 e |
| | Purslane | 73 a | 66 b | 54 c | 48 d | 39 e |
| | Amaranth | 65 a | 59 b | 53 c | 48 d | 37 e |

Means followed by the same letter within each column are not significantly different, as indicated by Tukey Test.