

# Biostimulation effects of rosemary essential oil on growth and nutrient uptake of tomato seedlings

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## ABSTRACT

Plant essential oils represent natural compounds that have a wide range of biologically activities on life systems. Various effects of essential oils on plant growth have been shown; however, their effects on nutrient uptake have not been yet assayed. In this study, effects of two concentrations of rosemary essential oil was evaluated on growth characteristics and nutrient uptake of tomato seedlings in a relatively lime soil and under greenhouse conditions. Treatments were foliar spray of 500 or 1000 ppm oil, soil application of 500  $\mu\text{L oil.kg}^{-1}$  soil, and a no application control. The seedling growth was changed under essential oil applications particularly by 1000 ppm foliar spray. Foliar application of 1000 ppm essential oil reduced plant height, whereas it increased leaf SPAD value, shoot and root fresh weights, leaf soluble carbohydrates and nutrient concentration of nitrogen, potassium, magnesium, iron and zinc of leaves than control plants. Root fresh weight (but not other traits) was also higher under foliar application of 500 ppm oil or soil application of oil than control plants. The results indicate that rosemary oil has beneficial effects on nutrient uptake of tomato seedlings toward a better growth quality.

## 1. Introduction

Agriculture intensification and current crop production is widely dependant on relatively high application rates of chemical fertilizers that generally results in various environmental challenges (Souri, 2016), irreversible global climate change and loss in many ecosystem services (Canellas et al., 2015). The promotion of plant growth is known as biostimulation, and many compounds can act to promote general plant growth or a given trait of plants such as root length or root dry matter (Canellas et al., 2015; Russo and Berlyn, 1992). Therefore, sustainable approaches are needed regarding agricultural production, particularly fertilization practices (Souri, 2016; Sánchez-Sánchez et al., 2002; Russo and Berlyn, 1992).

In agriculture system, various compounds are known to have biostimulation effects on plant growth and productivity. Organic acids, amino acids as well as compounds such as humic acid, salicylic acid and polyamines are among those stimulants that their soil or foliar application generally affects many plant growth related traits (Souri, 2016; Canellas et al., 2015; Marschner, 2011). A group of well defined biostimulants are essential oils that are natural product of complex nature, generally isolated from aromatic plants, showing a wide range of biological activities in life systems (Bakkali et al., 2008; Figueiredo et al., 2008). They frequently are responsible for the distinctive odor of plants.

They are mainly known for their antioxidant and anti-inflammatory activities in food, cosmetic industries and human health (Bakkali et al., 2008; Jalali-Heravi et al., 2011). Huge preliminary works have been done to demonstrate the potential of essential oils as antioxidant, and for use as biological control against fungi and bacteria as well as against postharvest pathogens (Bakkali et al., 2008; Figueiredo et al., 2008; Nerio et al., 2010). Antimicrobial properties of essential oils, as safer alternatives of synthetic preservatives, have been of great interest for many years to be applied in food, cosmetic and pharmaceutical industries. In poultry production systems, several studies show that including essential oils in poultry diets can improve digestive enzyme activities, nutrients uptake and growth performance in broiler chickens leading to an improved feed conversion ratio (Brenes and Roura, 2010). However, their role on plant growth still remains unclear.

In recent years, there has been an increasing interest in application of natural substances in biological systems including crop production. The antimicrobial properties of essential oils have been known for many centuries. Rosemary (*Rosmarinus officinalis* L.) is a perennial herb from Lamiaceae family, with needle-like leaves full of fragrant as essential oil. Rosemary is native to the Mediterranean region and as a tolerant plant grows in many countries. The leaves of this plant are full of fragrant and high in iron, calcium, and vitamin B6 (Jalali-Heravi et al., 2011). On the other hand, its oil has a wide range of action in

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**Table 1**  
Physio-chemical properties of the soil used in the experiment.

Potassium (mg. kg <sup>-1</sup> )	Phosphorus (mg. kg <sup>-1</sup> )	Nitrogen %	Organic C (%)	pH	EC <sup>a</sup> (dS m <sup>-1</sup> )	Clay %	MgCO <sub>3</sub> (%)	CaCO <sub>3</sub> (%)	Soil texture
256	15.2	0.096	0.62	7.87	2.15	20	1.4	6.8	Loamy

<sup>a</sup> EC = electrical conductivity.

medicine and in human health programs (Jalali-Heravi et al., 2011). Application of rosemary essential oils has shown various biological effects on plants (Bakkali et al., 2008; Jalali-Heravi et al., 2011). In this study, spraying rosemary essential oil on leaves of tomato seedlings or through their root feeding is proposed that may affect the nutrient uptake and general plant growth.

## 2. Material and methods

### 2.1. Experimental setup

This study was conducted during spring of 2011 and under greenhouse conditions, at Faculty of Agriculture, Tarbiat Modares Uni., Tehran-Iran. About 1 kg dry soil was placed in black plastic pots, and physiochemical properties of the soil presented in Table 1. Five tomato seeds (*Lycopersicon esculentum* var Green Supper) were sown 1 cm deep in each pot, and two weeks after germination they were reduced to one plant per pot. The greenhouse temperature was  $27 \pm 5^\circ\text{C}$  with 75–80% humidity and light intensity of  $250 \mu\text{mole m}^{-2} \text{s}^{-1}$ . Pots were irrigated with tap water in a daily basis and to 80% soil field capacity.

### 2.2. Rosemary oil extraction

Fresh rosemary foliage were cut from plants grown in Faculty of Agriculture, Tarbiat. Modares Uni., Tehran-Iran. The foliage were gently washed with tap water and were dried in shade. Two hundred gram of dried materials were steam-distilled for 90 min in full glass apparatus, with 1.9% oil yield production. The rosemary oil extraction was performed for two hour, after four hour maceration in 500 ml water (Jalali-Heravi et al., 2011). The rosemary oil was separated via Clevenger, and oil was placed in dark glass bottles and in refrigerator for one week.

### 2.3. Treatment application

Treatments were various concentrations of rosemary essential oil as follow: foliar application of 500 or 1000 ppm, soil application of 500  $\mu\text{L}$  essential oil per kg soil, and a zero application as control. Four replications were included in which each pot containing one tomato seedling represented as one replication, and treatments were arranged in completely randomized design.

Foliar applications of oil were started three weeks after seedling emergence and for each concentration (500 or 1000 ppm) three sprays were performed with one week intervals. For this purpose, the exact amount of 500 or 1000  $\mu\text{L}$  oil was dissolved in one liter distilled water and sprays were performed using a portable sprayer at early in the morning, just one hour after sunrise (Dehnavard et al., 2017). Soil application of rosemary oil was also done in relatively similar way, after addition of rosemary oil to distilled water that was applied to pots during growth period based on 80% soil field capacity. A final concentration of 500  $\mu\text{L kg}^{-1}$  soil was applied in three split applications including; the first 200  $\mu\text{L kg}^{-1}$  soil was applied three weeks after emergence, and two other applications of 150  $\mu\text{L kg}^{-1}$  soil were done in one week intervals.

### 2.4. Measurements

Plants were harvested after eight weeks after emergence in which

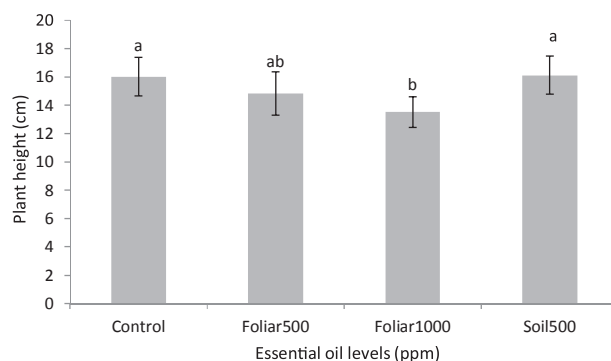
the final records of morphophysiological traits were presented in the results. Just before final harvest, plant height (as cm) was recorded using a ruler, and leaf SPAD values (The Soil and Plant Analysis Development) were recorded using SPAD meter (model 502 Plus, Illinois, USA). The average of 10 readings on different areas of plant leaves was presented. Plant shoots were cut at soil surface and shoot fresh weighs were recorded using a digital balance. For determination of root fresh weight the soils were gently washed out from the roots, and the fresh weight of collected roots after dewatering against tissue paper was measured using a digital balance. The soluble carbohydrates of leaves were measured using anthrone method, in which 0.1 g of leaf fresh tissues was extracted in 2.5 mL ethanol 80% at  $95^\circ\text{C}$  for 60 min (Fahimi et al., 2016). The extract was filtered and the alcohol was removed by evaporation. The anthrone reagent was used for preparation of samples and their absorbance was measured at 625 nm using a spectrophotometer. A standard glucose curve was also used for calculation of carbohydrate content. Leaf nutrient concentrations were determined with different apparatus. Nitrogen concentration of leaves was determined by the Kjeldahl method, K using flame photometry, and Mg, Fe and Zn were determined using atomic absorption spectrophotometer following (Souri and Roemheld, 2009).

### 2.5. Statistical analysis

Excel software was used for calculating means and standard deviations and data were analyzed by SPSS software. Comparison of means was performed at 5% by LSD test

## 3. Results and discussion

The result showed that tomato plant height (Fig. 1) was differently affected by essential oil treatments. Foliar spray of 1000 ppm but not 500 ppm oil has resulted in significantly shorter plants than control. Determination of plant leaf SPAD values (Fig. 2) showed that plants treated with foliar spray of 1000 ppm oil had the highest SPAD value (or chlorophyll index) that showed significant increase compared to control plants. Foliar application of 500 ppm oil and soil applied of



**Fig. 1.** The height of tomato seedlings affected by foliar and soil application of rosemary essential oil. Foliar spray of oil (in each level) was applied three times in one week intervals, during third, fourth and fifth week of growth out of eight weeks seedling growth period. Soil application of oil was in three split applications of a final volume of 500  $\mu\text{L/kg}$  soil. Data are average of four replications, and columns with at least one common letter are not significantly different at 5% level of LSD test.

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