Quantitative and qualitative performance of *Froriepia subpinnata* as affected by mycorrhizal symbiosis, compost tea, and vermicompost

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Abstract

This research involved in and coordinated multidisciplinary and complementary natural resources in the area of sustainable soil and plant nutrient management as well as economic benefits of edible herb production systems. The main concern was selection of the right growing media to achieve optimum yield and growth rate while avoiding application of synthetic fertilizers. *Froriepia subpinnata* is a medicinal and edible endemic vegetable which grows naturally in the north of Iran. In order to evaluate the effect of compost tea, vermicompost, and mycorrhizal symbiosis on morphophysiological traits of *Froriepia subpinnata*, a factorial experiment was conducted in Sari, Mazandaran based on randomized complete block design with four replications. The factors included *Glomus moseae* at two levels (inoculated and non-inoculated), vermicompost at two levels (without application and application at 2 tons/ha), and compost tea at two levels (no application and application at 1.5 liter/ha). Results showed that the impact of dual interactions of vermicompost and mycorrhizal symbiosis were significant on the length and diameter of flowering stems, flower diameter, leaf width, and plant fresh weight at p < 0.01. The triple interactions of the factors on leaf area, plant dry weight, and essential oil yield were significant at p < 0.01. The highest plant dry weight (0.81 g/plant) and the highest essential oil content % 0.82 were observed with application of vermicompost and mycorrhizal symbiosis.

Keywords: Apiaceae; aromatic vegetable; endemic herb; foliar application; *Froriepia subpinnata*


Introduction

Native and endemic plants, wild species, and natural vegetation are considered as plant genetic resources of each region and are the main elements of biodiversity. Plant genetic resources provide livelihood resources such as food, grazing, fuel, fiber, medicine, and many other human needs. Due to the different morphological and physiological characteristics and adaptation to ecological conditions of each region, endemic plants have important effects on ecosystem dynamics. *Froriepia subpinnata* has 150 species in Eurasia and Africa and more than 16 species in Europe. *Froriepia subpinnata* grows wildly in Central, West and North of Iran and especially in mountainous areas with cold climates (Jodral, 2004). In traditional medicine,
this plant has been used as a carminative, appetizer, antiseptic, antispasmodic, diuretic and sedative in order to increase the milk secretion. *Froriepia subpinnata* contains antioxidant and antibacterial properties (Sivam, 2001; Gülçın et al., 2003) and essential oils of 39 species have been identified as anti-cancer compounds. Consuming this plant as a flavoring agent in food industry is affordable due to its naturally growing property and its leaves and stems are used in the preparation of local dishes. Considering the importance of medicinal plants in various industries, these plants are producing without application of harmful chemical inputs. Proper management and integration of symbiotic microbial species with medicinal plants affect their quality and yield (Abudul-Jaleel et al., 2007). In addition to the importance of medicinal and aromatic plants for medical uses, they have been used extensively in many fields related to the food industry, cosmetics, health, and food (Jesus, 2003).

Mycorrhizal symbiosis has a symbiotic relationship with the roots of most crops and increases the absorption of water and nutrients such as phosphorus and some micronutrients, reduces negative impacts of environmental stress, increases resistance to pathogens, and improves the plant growth and performance of host plants in sustainable agricultural systems (Sharma, 2002).

Vermicompost which is produced by earthworms resulting from the transformation and partial digestion of the organic waste (manure, crop residues, etc.) while passing through the digestive system of worms. Vermicompost has high porosity, high level of available nutrients with long term storage, providing ventilation and proper drainage, high water holding capacity, positive effects in increase population and activity of beneficial soil microorganisms (such as mycorrhiza fungi and phosphate solubilizing microorganisms), and in general improves the crops growth and yield. Arancon et al. (2004) stated that sustainable agriculture increases the crop quality and yield stability. The medicinal herbs which are considered as healthy products are suitable for this system (Gupta et al., 2002). Mycorrhizal symbiosis has a beneficial impact in increasing the ability of the host plant for non-mobile nutrient uptake, especially phosphorus and several other micronutrients. Copetta et al. (2006) reported that the inoculation of *Ocimum basilicum* L. with three mycorrhiza fungi species of *Gigaspora rosea* BEG 9, *Glomus mossae* BEG 12, and *Gigaspora margarita* BEG 34, significantly increased plant height, leaf number and surface area, biomass, root length and lateral growth, and essential oil content compared with control treatment. It was also found that the use of vermicompost increased the yield of two species of plantain (*Plantago major*) as a medicinal plant (Sanchez et al., 2008). In a survey on *Matricaria chamomilla*, it was observed that using vermicompost significantly increased the plant height and plant yield (Azizi et al., 2008). Vermiwash, as the vermicompost extract, is a set of substances and waste from earthworms combined with major micro-nutrients and soil organic molecules used for plant development and growth and applied as foliar spray. Plant spraying by vermiwash stimulates the growth and increases the crop yields, causing resistance against a variety of environmental factors. This substance can prevent damages to plants such as leaf blight (Sivasubramanian and Ganeshkumar, 2004; Quaik et al., 2012).

This study aimed at investigating the effect of organic based fertilizers and mycorrhizal symbiosis on the content of nutrients and some morphophysiological characteristics of *Froriepia subpinnata* in Sari- Mazandaran Province in north of Iran.

**Materials and Methods**
Froriepia subpinnata under mycorrhizal symbiosis, compost tea, and vermicompost

The study was conducted in 2014 in Sari (Mazandaran Province), in north of Iran (53° 63' E, 36° 82' N) using a factorial experiment in the form of randomized complete block design with four replications. The factors included *Glomus mosseae* at 2 levels (no application and application at 2 tons/ha), vermicompost at 2 levels (no application and application at 2 tons/ha), and compost tea at 2 levels (no application and application at 1.5 liters/ha). Compost tea was sprayed at 4-5 leaf stage on the target plots. The leaf area was measured by Image J Software. Measurements were conducted at two levels of vegetative stage (before flowering) and reproductive stage (after flowering). The size of each plot was 1.5 × 1.5 square meter and six rows of planting were considered in each plot. Results of soil and organic fertilizer analysis are shown in Tables 1 and 2, respectively.

Planting operations were conducted in November, 2014, which involved irrigation practices, thinning, and weeding. The irrigation was performed once every 4 days after ensuring the plant growth and establishment. In order to measure some morphological features and essential oil content, 8 plants per plot were randomly harvested and the height and lateral branches were measured and recorded separately. At maturity stage, the plant characteristics such as dry weight, number of seeds per plant, and 1000 seeds weight were measured. Twenty grams of dried samples were mixed with distilled water in a Clevenger apparatus to extract the essential oil. The essential oil extraction time for all samples was 3 hours. After dehumidifying the oil and using sodium sulfate, the oil content was determined based on the percentage.

Collected leaves were dried and powdered by an electric mill and underwent the digestion process using sulfuric acid, salicylic acid, hydrogen peroxide, and selenium to prepare the extracts and determine the nutrient concentrations. Nitrogen content estimated using titration method after distillation in Kjeltec Auto Analyzer (Bremner and Mulvaney, 1982). Phosphorus content was analyzed using colorimetric method (yellow molybdate vanadate). Trace elements such as iron, copper, zinc, manganese, and potassium content were also measured using spectrophotometry and atomic absorption. Potassium content was analyzed using the flame emission through a flame photometer. Analysis of variance was done using SAS statistical software and means were compared using the Duncan’s Multiple Range Test.

### Results

**Yield components**

According to the results of data analysis (Tables 3 and 4), the combined and single use of vermicompost and mycorrhizal symbiosis had significant effects on shoot length, number of leaves, leaf surface area, stem diameter, and leaf length and width (p<0.01), but the application of compost tea had no significant effect on the leaf area and width (Tables 3 and 4). Mean comparisons showed that the application of

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**Table 1**

<table>
<thead>
<tr>
<th>Texture</th>
<th>OC (%)</th>
<th>Potassium (ppm)</th>
<th>Total Nitrogen (%)</th>
<th>Phosphorus (ppm)</th>
<th>OM (%)</th>
<th>T.N.V (%)</th>
<th>pH Paste</th>
<th>EC (ds.m⁻¹)</th>
<th>depth (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>L</td>
<td>2.9</td>
<td>296</td>
<td>0.20</td>
<td>5.3</td>
<td>3.27</td>
<td>27</td>
<td>7.63</td>
<td>0.54</td>
<td>0-30</td>
</tr>
</tbody>
</table>

**Table 2**

<table>
<thead>
<tr>
<th>Copper (ppm)</th>
<th>Manganese (ppm)</th>
<th>Zinc (ppm)</th>
<th>Iron (ppm)</th>
<th>Magnesium (%)</th>
<th>Calcium (%)</th>
<th>Phosphorus (%)</th>
<th>Potassium (%)</th>
<th>Nitrogen (%)</th>
<th>OM (%)</th>
<th>Ec</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>22</td>
<td>79.3</td>
<td>266</td>
<td>1981</td>
<td>0.15</td>
<td>4.09</td>
<td>2.2</td>
<td>3.9</td>
<td>1.55</td>
<td>20.17</td>
<td>1.2</td>
<td>6.35</td>
</tr>
</tbody>
</table>
vermicompost and mycorrhizal symbiosis caused a significant increase in stem length and leaf number compared to the control. The highest number of leaves/plant (25.92), the maximum length of the stem (18.77 cm), the highest number of stems/plant (18.70), the maximum leaf area (66.21 cm), and the maximum leaf length (1.22 cm) (Table 8) were obtained in the treatments with no mycorrhiza application while compost tea and vermicomposting were applied; and the maximum leaf width (0.70 cm) was achieved in the treatment without application of vermicompost (Table 7).

**Length and diameter of flowering stem**

The variance analysis (Table 3) showed that the application of compost tea in combination with mycorrhizal symbiosis had a significant effect on the length and diameter of the flowering stem (p<0.01). Double interaction of vermicompost and mycorrhizal symbiosis showed the highest flowering stem diameter and length in the treatments without vermicompost (Table 7) and (4.53 mm for stem length) in the treatments with mycorrhizal symbiosis while no compost tea was applied (Table 6).

**Number of flowers and flower diameter**

The analysis of variance (Table 3) showed that the main effect, double and triple interactions of application of vermicompost, compost tea, and mycorrhizal symbiosis had significant effects on flower number and flower diameter (p<0.01), but compost tea and triple interactions of the factors did not have any significant effects on the flower diameter (Table 3). Results of mean comparisons showed that combined application of vermicompost and mycorrhizal symbiosis increased the number of flowers and flower diameter compared to the control. The highest number of flowers was achieved (83.27/plant) when vermicompost and mycorrhiza fungi were applied together (Table 8), and the maximum diameter of the flower (1.41
mm) was obtained while both vermicompost and mycorrhiza fungi were applied (Table 7).

Plants fresh and dry weight

The variance analysis showed that the main effect and double interactions of application of mycorrhiza fungi and vermicompost were significant on fresh and dry weight of plants (p<0.01) but the application of vermicompost, double interactions of compost tea and vermicompost, and triple interactions had no effect on plant dry and fresh weights (Table 3). Mean comparisons showed that the application of vermicompost and mycorrhizal symbiosis caused an increase in plant fresh and dry weight compared with control treatments in which the highest dry weight (0.81 g/plant) was attributed to the vermicompost and mycorrhizal symbiosis treatments without compost tea (Table 8). The highest plant fresh weight (4.62 g/plant) was related to the mycorrhizal symbiosis treatment without compost tea (Table 6).

Number of seeds per plant and 1000 seeds

The results of variance analysis (Table 3) revealed that the main, dual, and triple interactions of mycorrhizal symbiosis, vermicompost, and compost tea had a significant effect on the number of seeds/plant at p<0.01 level, but application of compost tea, double interactions of compost tea x vermicompost, and triple interactions had no effect on 1000 seeds weight (Table 3). The highest number of seeds/plant (163.55) was recorded in the treatments containing mycorrhizal fungi applied with compost tea and vermicompost (Table 8).

Root parameters
Variance analysis (Table 4) showed that the main and dual interactions of applying vermicompost and mycorrhizal symbiosis had significant effect on root length, root dry weight, and root diameter at p<0.01 level, but the double interactions of compost tea and vermicompost had no effect on root dry weight. The comparison of the triple interactions showed the highest root diameter (4.87 mm) and length (8.24 cm) which were achieved in treatments with mycorrhizal symbiosis and vermicompost without compost tea (Table 8) and highest roots dry weight (3.49 g/plant) which was obtained in the application of vermicompost and mycorrhizal symbiosis without compost tea (Table 8).

**Essential oil content and essential oil yield**

The results of variance analysis showed that the application of mycorrhiza fungi, compost tea, and vermicompost increased the essential oil content and essential oil yield compared to the control treatments and their interaction was significant at p<0.01. The highest percentage of essential oil contents was 0.82 when the plants were inoculated with mycorrhizal fungi and vermicompost without application of compost tea was 0.26 (Table 8).

**Leaf yield (dry)**
Variance analysis (Table 4) showed that the combined and single application of vermicompost and mycorrhiza fungi had significant effects on *Froriepia subpinnata* dry leaf yield (*p*<0.01). The mean comparisons showed that the highest dry leaf yield was achieved through mycorrhizal symbiosis treatment (1564.9 kg/ha), while the lowest accompanied by the application of compost tea and vermicompost (422.3 kg/ha) (Table 8).

**Plant nutrient contents**

Table 10
Mean comparison of the main effects of vermicompost, compost tea, and mycorrhizal symbiosis on nutrient contents of *Froriepia subpinnata*

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Nitrogen (%)</th>
<th>Phosphorous (%)</th>
<th>Potassium (%)</th>
<th>Copper (ppm)</th>
<th>Zinc (ppm)</th>
<th>Manganese (ppm)</th>
<th>Iron (ppm)</th>
<th>Mansesium (%)</th>
<th>Calcium (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>V</td>
<td>0.62c</td>
<td>0.13c</td>
<td>3.82b</td>
<td>20.75a</td>
<td>11.2c</td>
<td>61a</td>
<td>622a</td>
<td>0.68b</td>
<td>1.10b</td>
</tr>
<tr>
<td>T</td>
<td>2.33a</td>
<td>0.16b</td>
<td>4.32a</td>
<td>10.25c</td>
<td>17b</td>
<td>25.7b</td>
<td>363 c</td>
<td>1.02a</td>
<td>1.09b</td>
</tr>
<tr>
<td>M</td>
<td>1.90b</td>
<td>0.18a</td>
<td>3.25c</td>
<td>12.75b</td>
<td>30.7a</td>
<td>25b</td>
<td>394.5b</td>
<td>0.39c</td>
<td>2.67a</td>
</tr>
<tr>
<td>Control</td>
<td>0.57d</td>
<td>0.11d</td>
<td>0.81d</td>
<td>8.66d</td>
<td>7.3d</td>
<td>17.3c</td>
<td>134d</td>
<td>0.26d</td>
<td>1.05c</td>
</tr>
</tbody>
</table>

(T): compost tea, (V): vermicompost, (M): mycorrhizal symbiosis

Results of variance analysis (Table 9) showed that different treatments had a significant effect on *Froriepia subpinnata* nutrients (*p*<0.01). Mean comparisons showed that the application of vermicompost and mycorrhizal symbiosis significantly increased the nutrients in *Froriepia subpinnata*. The highest (%2.32) and lowest (%0.57) levels of nitrogen were achieved with application of compost tea and the control treatments, respectively. The highest amount of phosphorus was achieved in the treatments with mycorrhizal symbiosis (18%) while the lowest amount with control treatment (11%). The highest and lowest potassium levels were observed in compost tea (%64.32) and control treatments (%0.81), respectively (Table 10) which was similar with the results of Zaefarian et al. (2013) and Ardakani et al. (2011). The maximum amounts of copper (20.75 ppm), Mn (61 ppm), and Fe (622 ppm) were achieved with application of vermicompost. On the other hand, the highest amounts of Zn (30.7 ppm) and calcium (2.67%) were obtained using mycorrhiza fungi. Finally, the highest amount of magnesium (1.02%) was obtained in the treatments involving application of compost tea.

**Discussion**

The results showed that mycorrhizal symbiosis has a positive effect on plant leaf area which is similar in many other edible plants (Amiri et al, 2013). Moradi et al. (2009) reported the pea plant height increased by Glomus inoculation. Widada et al. (2007) stated that *Sorghum bicolor* inoculated with mycorrhiza had a higher plant height. Koozehgar Kaleji (2014) reported that the use of biological fertilizers significantly increased the diameter of flowering stem of *Eryngium caeruleum*. Tahami Zaranadi et al. (2010) reported a positive effect of organic and biological fertilizers on increasing number of flowering stems and the number of flowering cycles in *Ocimum basilicum*. Increasing the number of flowers in this study was attributed to the improvement of the water and nutrients absorption by plants which were inoculated with mycorrhiza fungi. The application of vermicompost increased the number of flowers per plant for *Echinacea purpurea* (Razvinia et al., 2015). Taher et al. (2013) reported that the effect of solubilizing phosphate bacteria on Tuberose flower diameter increased significantly. Bigonah et al. (2014) stated that biological fertilizers and
vermicompost increased plant dry weight in cilantro. Farzaneh et al. (2009) reported that the seed inoculation of peas with mycorrhiza fungi increased total dry weight (43%) compared with the control treatments. The reason for such an increase was ascribed to the length of root hairs and fungal hyphae. Bigonah et al. (2015) stated that biological fertilizer and vermicompost increased seeds yield per plant in cilantro. In other experiments, the application of organic and biological fertilizers had relatively similar effects on the total weight of seeds per plant and 1000 seeds weight in Ocimum basilicum L. (Tahami Zarandi et al., 2010) and fennel (Moradi 2009). Abrishamchi et al. (2014) stated that the application of vermicompost significantly increased tomato root diameter. Sirrenberg et al. (2007) reported that in plants inoculated with mycorrhiza fungi, auxin synthesis increased significantly which had a positive effect on root growth. Production of plant hormones (plant growth substances) and increase in enzymes activities by mycorrhizal symbiosis can intensify the plant and root growth leading to higher absorption of water and nutrients. Samiran et al. (2010) stated that the bean root in the presence of vermicompost increased significantly. Azimi et al. (2013) reported that the thyme inoculated with mycorrhizal fungi (G. intraradices and G. mosseae) species increased root dry weight compared to the control treatments. Abrishamchi et al. (2014) reported that the application of vermicompost significantly increased root dry weight of tomato. Mycorrhizal symbiosis also can enhance root development in alfalfa as reported by Ardakani et al. (2009). Kapoor et al. (2004) stated that fennel root symbiosis with two species of vesicular-arbuscular mycorrhiza enhanced essential oil content and quality. Darzi et al. (2009) reported that vermicompost increased the percentage of essential oil in fennel. Mona et al. (2008) reported that vermicompost positively affected fennel essential oil. Considering the fact that the essential oil function is part of the dry weight and essential oil percentage, any increase in these two cases can lead to increased oil yield. The results of many other experiments showed that mycorrhizal symbiosis increased photosynthetic rate by increasing the leaf area, accordingly (Valentine et al. 2006). Azizi et al. (2005) investigated the positive effects of different levels of vermicompost to improve uptake of nitrogen, phosphorus, and potassium and stated that vermicompost and vermiwash were effective on the amount of Ocimum basilicum L. active ingredients. Compost tea contains humic acid which improves the availability of certain nutrients, especially iron and zinc. The main factor for increasing availability of organic waste consumption is the formation of organic complexes (Atiye et al., 2002).

Conclusion

Results show that, the application of mycorrhiza fungi, compost tea and vermicompost had significant increase in the measured traits. In fact, increasing yield and its components when using mycorrhiza fungi and the application of vermicompost and compost tea when spraying on the plant caused increased absorption of nutrients such as nitrogen, phosphorus, potassium and micronutrients. Since the mycorrhizal symbiosis had a positive effect on the plants, they can be used as a substitute for chemical fertilizers in order to move towards sustainable agricultural production.

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