



Studying the quantitative and qualitative characteristics of vetiver grass (*Chrysopogon zizanioides* L.) under different compost and zeolite treatments

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Abstract

Vetiver grass (*Chrysopogon zizanioides* L.) is a multi-purpose plant which plays important role in soil conservation due to its deep roots. Since the soil in most regions of Iran is poor in organic matter, it is necessary to apply proper soil amendments to optimize plant growth. A completely randomized factorial experiment with three replications was done in the research greenhouse of Malayer University to evaluate the combined effect of zeolite and compost on quantitative (i.e. shoot dry weight and root dry weight) and qualitative (i.e. chlorophyll a and b, carotenoids, relative water content, and proline) characteristics of Vetiver grass. Compost was added at four levels of zero (control), 20, 40, and 60 percent by volume and zeolite was added at five levels of zero (control), 10, 20, 30, and 40 g to the soil (five kg) of the pots containing scions. The results demonstrated that the applied treatments had a significant effect on all the measured traits. The concentrations of chlorophyll a and b and carotenoids significantly increased ($p \leq 0.05$) under the effect of different levels of compost and zeolite. Although the leaf relative water content (RWC) increased in some of the treatments (Z2C3, Z2C4, Z4C1, and Z5C3), in general, it decreased significantly compared to the control treatment. In addition, the shoot and root dry weights increased significantly under the effect of all fertilizer levels. In our study applying fertilizers had a positive effect on the quantitative and qualitative performance of Vetiver grass.

Keywords: Clinoptilolite, Improvement, Fertilizers, Rangeland, Compost

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Introduction

Vetiver grass, which is known today by the scientific name *Chrysopogon zizanioides* L., is a herbaceous and perennial plant and belongs to the family of Poaceae (Chahal et al., 2015). Vetiver grass is a tropical plant native to India and has a high adaptability to different environmental conditions. Its height reaches 150 to 200 cm and its root length is 3 to 4 meters (Niknahad et al., 2014). The deep roots of this plant cause more water penetration and increase soil moisture accessibility for other plants (Du and Truong, 2000).

The main reason for using Vetiver grass is it has deep and wide roots and such it is good for soil and water conservation in a wide range of different cropping patterns. In addition, all parts of Vetiver grass can be used. For instance, it can be used for forage production (Matew et al., 2016) and mushroom cultivation (as a substrate). It can also be used for water treatment, weed control, perfume production, medicinal usage, and handicraft production (Sornpraserta and Aroonsrimorakotb, 2014). In addition, the essence and fragrance of Vetiver grass can be used as a new strategy for controlling vermin including termites (Ewetola et al., 2017).

Soil erosion as one of the fundamental problems of rangelands can be significantly controlled by planting and improving the growth of Vetiver grass hedges (Rao et al., 2015). Although Vetiver grass is able to grow in different environmental conditions, it is essential to know its reactions to fertilizers (such as zeolite and organic fertilizers) and the changes in its quantitative and qualitative characteristics. Research shows that using organic fertilizers especially in the form of compost not only increases the performance of the plant but also has positive and beneficial effects on the physicochemical properties of soil leading in turn to the health and fertility of the soil and to a sustainable production in the long term (Carmo et al., 2016).

In arid and semi-arid regions where the soil mainly has a low fertility, in addition to increasing the organic matter of soil, Vetiver grass can decrease acidity and supply the plants with nutrients (Malakuti,

1996). Zeolites are hydrated aluminum-alkaline and alkaline-earth minerals (Tsintskaladze et al., 2016). Clinoptilolite is the most abundant zeolite (Erdem et al. 2004) with the chemical formula $(Al_8SiO_{40}O_{96}) 24H_2O (Na_4K_4)$. Due to its high exchange capacity, high water retention capacity (Hosseini Abari et al., 2007), environmental adaptability, cost-effectiveness (Abbohanna and Salam-Titinchi, 2009), and remarkable abundance in Iran (Kazemian, 2000), this mineral is suitable for soil amendment.

Zeolites improve soil conditions, increase soil moisture retention capacity, prevent soil moisture loss, and satisfy the needs of plants during their growth period by gradually releasing nutrients (Mumpton and Ormsby, 1987). Since the soil in most regions of the country is poor in organic materials, the use of fertilizers (especially harmless non-chemical ones) not only improves economic performance but also is effective in reducing environmental pollutions.

Despite the existence of rich zeolite resources in Iran, its use for rangeland plants has rarely been studied. Therefore, in this study, the interaction effect of this mineral with compost manure on the quantitative (dry weight and length of roots and shoots) and qualitative (chlorophyll a and b, carotenoid, relative humidity, and proline) properties of Vetiver grass has been investigated in the research greenhouse of Malayer University.

The dry weight and length of the shoot indicate the amount of forage production and forage palatability, respectively. The root length and weight are important for soil conservation. Chlorophyll a and b, carotenoid, relative humidity, and proline are the parameters indicating the amount of forage palatability. We have chosen to focus on these properties in the current research.

Materials and Methods

This study was implemented in the spring of 2016 in the research greenhouse of the Faculty of Natural Resources and Environment of Malayer University located at the eastern longitude of 48° 49', northern

latitude of 34° 17', and a height of 1780 m above sea level. This research was carried out in pots in a completely randomized factorial design with three replications.

Clinoptilolite zeolite (with the particle size of 0.1 to 1.5 mm and the price of 700 rials per kg) was purchased from Afrazand Company, Tehran. Vetiver scions obtained from Ramhormoz were maintained in the greenhouse for five months between 2015 and 2016 to adapt to the new environmental conditions.

The scions of Vetiver were placed in 60 five-kg plastic pots with a height of 20 cm and diameter of 16 cm in a tripod manner. Zeolite fertilizer was added at five levels of zero (control), 10, 20, 30, and 40 g to the soil (five kg) of the pots and mixed with soil and manure compost at four levels of zero (control), 20, 40, and 60 percent by volume. Zeolite fertilizer was mixed with soil thoroughly as it is not leached. However, the manure compost was added to the first 15 cm of the pot soil because it is leached.

To measure the leaf relative water content (RWC), sampling was done on the last mature leaf and the samples were then immediately placed in plastic bags and transferred to the laboratory. After measuring the weight of fresh leaves, the samples were immersed in distilled water for 24 hours and the turgid weight of the leaves was recorded.

The samples were placed in an oven for 48 hours at 70°C and then their dry weight was measured. Using the following equation, the RWC of the leaves was calculated (Dhopte and Manuel, 2002):

$$\text{RWC}\% = (\text{FW}-\text{DW}/\text{TW}-\text{DW}) \times 100 \quad (1)$$

Where FW, DW, and TW are leaf fresh weight, leaf dry weight, and leaf turgid weight, respectively.

Chlorophyll a and b and the carotenoid content were measured using Arnon's method (Arnon, 1949). The method of Bates et al. (1999) was used to measure the proline content. After 6 months, the length of the roots was measured after they were cut off from the shoots in the laboratory. The leaves and roots were separately put in paper bags and placed in the oven for 72 h at 80 °C (Rahimi et al. 1389) and then their weights were measured.

Finally, the data were analyzed using SAS software (version 9.1) and the average-to-average comparison method. The differences between means were tested for significance by Duncan's multiple range test at $p \leq 0.05$.

Results

Concentration of chlorophyll a

Based on the results, different fertilizer levels had a significant impact on the concentration of chlorophyll a in *Chrysopogon zizanioides* ($P \leq 0.05$) (Figure 1). The most increasing effect was observed at the levels of 10 and 40 g of zeolite (Z2C1 and Z5C1, respectively) with 3.7 mg g⁻¹ leaf dry weight which did not have a significant difference with the treatment of 20 g of zeolite and 20 percent by volume of compost (Z3C2). The least increasing effect was observed in the treatment of 20 g of zeolite and 60 percent by volume of compost (Z3C4) with 1.3 mg g⁻¹ dry weight which did not show a significant difference with the control treatment. In addition, applying the highest level of fertilizer (Z5C4) did not have a significant difference with the control treatment (Figure 1).

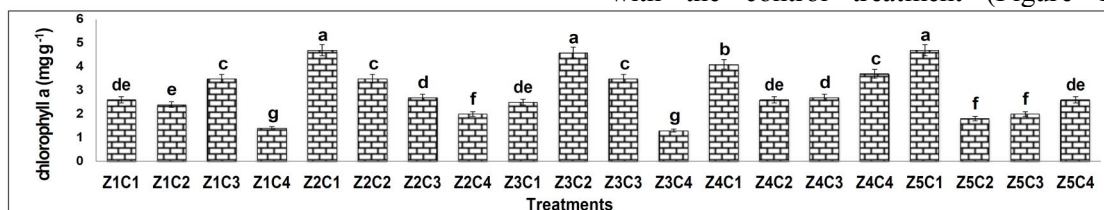


Figure 1. Changes in the concentration of chlorophyll a under different zeolite and compost treatments in *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the levels of 0, 10, 20, 30, and 40 g kg⁻¹ of zeolite, respectively, and letters C1, C2, C3, and C4 represent the levels of 0, 20, 40, and 60 percent by volume of compost)

Concentration of chlorophyll b

In total, the concentration of chlorophyll b in all the treatments was lower than that of chlorophyll a. The effect of fertilizer levels on chlorophyll b was similar to that of chlorophyll a with a slight difference (Figure 2).

The highest concentration of chlorophyll b was observed in the absence of compost

and 10 g of zeolite (Z2C1) treatment which was 61.53% more than that of the control treatment. The lowest concentration was observed in the treatment of 20 g of zeolite and 60 percent by volume of compost (Z3C4) which was 46.6% less than that of the control treatment indicating a significant difference in the concentration of chlorophyll b at different fertilizer levels (Figure 2).

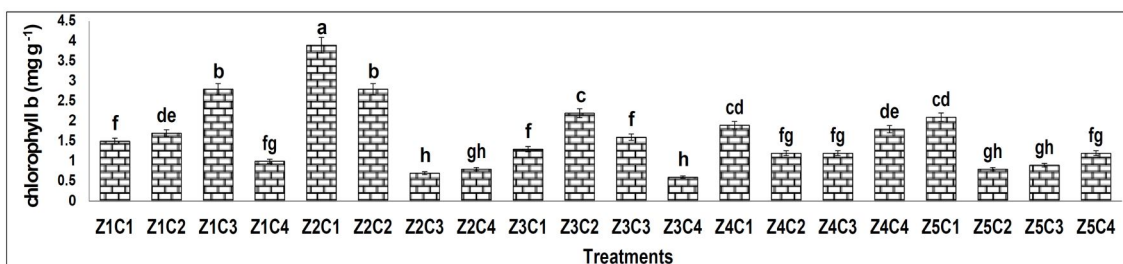


Figure 2. Changes in the concentration of chlorophyll b at different levels of zeolite and compost fertilizers in the plant species *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the control levels of 0, 10, 20, 30, and 40 gram per kilogram of zeolite, respectively, and letters C1, C2, C3, and C4 represent the control surfaces of 0, 20, 40, and 60 percent by volume of compost)

Concentration of carotenoids

The highest carotenoid concentration was observed in the treatment of 20 g of zeolite and 20 percent by volume of compost (Z3C2) and the lowest carotenoid concentration was observed in the absence of zeolite and 60 percent by volume of

compost and 20 g Kg⁻¹ of zeolite and 60 percent by volume of compost (Z1C4 and Z3C4, respectively) treatments. The changes of carotenoids with different fertilizer treatments were similar to those of chlorophyll a and b (Figure 3).

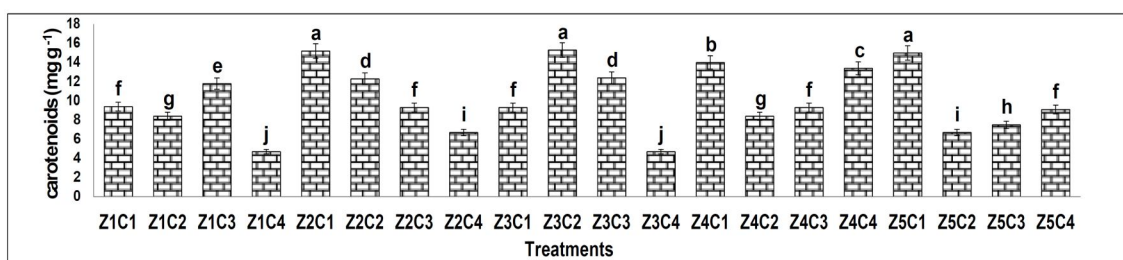


Figure 3. Changes in the concentration of carotenoids at different levels of zeolite and compost fertilizers in the plant species *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the control levels of 0, 10, 20, 30, and 40 gram per kilogram of zeolite, respectively, and letters C1, C2, C3, and C4 represent the control surfaces of 0, 20, 40, 60 percent by volume of compost)

Relative water content (RWC)

Although there was no significant difference among most of the treatments, in general, the applied fertilizers had a significant decreasing effect on the relative water content ($P \leq 0.05$).

The highest percentage of RWC was observed in the treatment of 30 g of zeolite

and the absence of compost (Z4C1) which had no significant difference with the control treatment, while the lowest percentage of RWC was observed in the treatment of 60 percent by volume of compost and the absence of zeolite (Z1C4) (Figure 4).

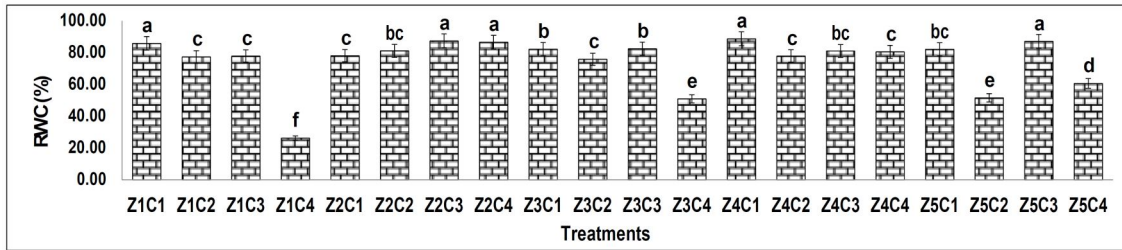


Figure 4. RWC changes at different levels of zeolite and compost fertilizers in the plant species *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the control levels of 0, 10, 20, 30, and 40 gram per kilogram of zeolite, respectively, and letters C1, C2, C3, and C4 represent the control surfaces of 0, 20, 40, and 60 percent by volume of compost)

Concentration of proline

The results showed that different fertilizer levels did not lead to a significant difference. However, a significant increasing difference was observed in the concentration of proline in the leaves ($P \leq 0.05$). The highest proline content was

observed in the treatment of 30 g of zeolite and the absence of compost (Z4C1), while the lowest proline content was observed in the control treatment and the absence of zeolite and 60 percent by volume of compost (Z1C4) treatment (Figure 5).

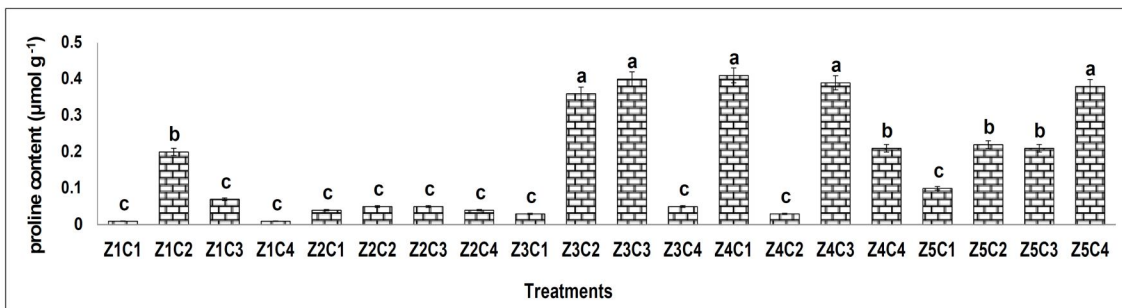


Figure 5. Changes in the concentration of proline at different levels of zeolite and compost fertilizers in the plant species *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the control levels of 0, 10, 20, 30, and 40 gram per kilogram of zeolite, respectively, and letters C1, C2, C3, and C4 represent the control surfaces of 0, 20, 40, and 60 percent by volume of compost)

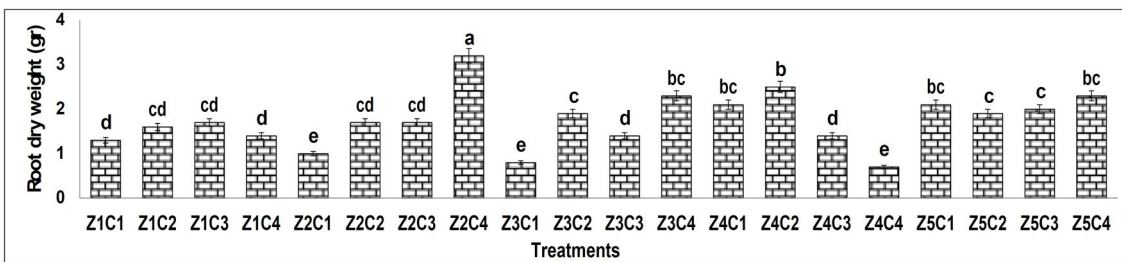


Figure 6. Root dry weight changes at different levels of zeolite and compost fertilizers in the plant species *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the control levels of 0, 10, 20, 30, and 40 gram per kilogram of zeolite, respectively, and letters C1, C2, C3, and C4 represent the control surfaces of 0, 20, 40, and 60 percent by volume of compost)

Root dry weight

The root dry weight was significantly influenced by different fertilizer levels

($P \leq 0.05$). The statistical analysis showed that the highest root dry weight belonged to the treatment of 10 g of zeolite and 80

percent by volume of compost (Z2C4). This indicated an increase of 59.37 percent compared to the control and 78.13 percent compared to the lowest amount. The lowest amount belonged to the treatment of 30 g of zeolite and 60 percent by volume of compost (Z4C4) which decreased 53.84% compared to the control treatment (Figure 6).

Shoot dry weight

The difference in the performance of shoot dry weight in different fertilizer treatments was significant so that the highest shoot dry weight was observed in the absence of zeolite and 40 percent by volume of compost (Z1C3) treatment while the lowest shoot dry weight belonged to the lack of zeolite and 60 percent by volume of compost (Z1C4) treatment (Figure 7).

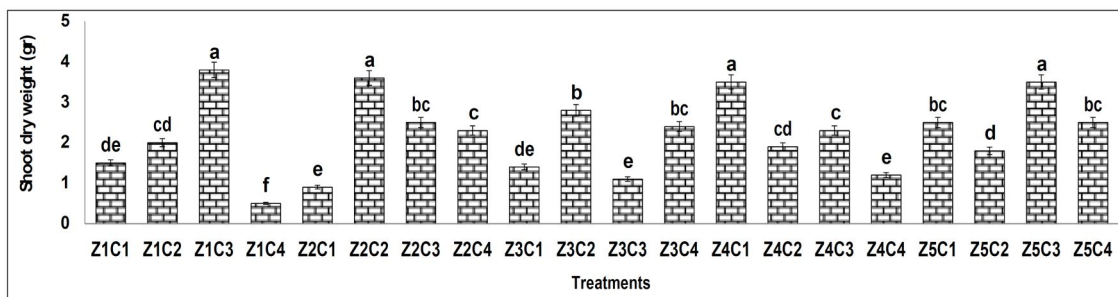


Figure 7. Shoot dry weight changes at different levels of zeolite and compost fertilizers in the plant species *Chrysopogon zizanioides* (dissimilar latin letters indicate significant differences of means in different treatments. Letters Z1, Z2, Z3, Z4, and Z5 represent the control levels of 0, 10, 20, 30, and 40 gram per kilogram of zeolite, respectively, and letters C1, C2, C3, and C4 represent the control surfaces of 0, 20, 40, and 60 percent by volume of compost)

Discussion

Concentration of chlorophyll and carotenoid

The statistical analysis results showed that different levels of zeolite and compost fertilizer had a significant impact on the concentration of chlorophyll a and b and carotenoids in Vetiver plant. Although this trend was not linear and declined in some levels, overall, fertilizer treatments had a significant impact on the concentration of chlorophyll. The results of this experiment are in accordance with the study of Bahador et al. (2015) on *Vigna radita* L. and with that of Farahmand et al. (2007) on *Narcissus tazetta*.

Since chlorophyll has a nitrogenous structure, the increased amount of chlorophyll in zeolite treatments may be due to the reduced leaching of nitrogen and the improvement of soil conditions (Khan et al., 2009), resulting in better growth of the light-absorbing system and better seedling establishment (Bahador et al., 2015).

In this experiment, with the addition of zeolite, the chlorophyll content decreased at

some levels probably due to the absorption of some nitrogen by zeolite (Yang et al., 2003). However, in general, adding zeolite had a positive effect on chlorophyll concentration. Manure, especially in the form of compost, contains large amounts of organic materials which are easily broken down and have large amounts of nitrogen (Bauer and Black, 1994). The use of manure compost in our study led to different chlorophyll concentrations. However, in general, it had a positive impact on the concentration of chlorophyll.

Azarmjoo et al. (2009) stated that the use of compost had a positive effect on physiological parameters such as chlorophyll a and b and was effective in increasing the nutrients of soil and their absorbance by the plants. On the other hand, Pourmoosavi et al. (2007) reported that the use of manure could not make a significant difference in the amounts of chlorophyll a and b. In our study, there was no significant difference between some levels.

Relative Water Content

As can be seen in Figure 3, initially, in the

absence of zeolite, with increase in compost, the relative water content had a significant decline. However, at other fertilizer levels, with increase in zeolite fertilizer, the relative water content had a significant increase. A decline in the value of some treatments (Z3C4 and Z5C2) was observed.

In general, zeolite had a more positive effect on leaf RWC than compost. This may be due to the fact that zeolite prevents moisture from leaving the soil (Malakuti, 1996) since the relative water content is associated with the soil moisture (Khan et al., 2007). Naeemi et al. (2012) also stated that the use of zeolite increased the relative water content compared to control. In addition, increasing the compost manure at some levels (Z2C3 and Z2C4) led to an increase in relative water content. The reason for this is probably the characteristics of manure such as improved soil moisture retention which increases leaf relative water content. Rahbarian and Salehi Sardoie (2013) also stated that by increasing manure, the relative water content increased. In the present experiment, the interaction of compost and zeolite reduced the relative water content which may be due to the characteristics of both of these fertilizers in retaining moisture and gradually releasing it during plant growth. Given the short duration of the experiment, the applied fertilizers could not make a positive effect on the relative water content.

Proline content

Proline has nitrogenous compounds; hence, the increase of nitrogen content in soil increases the production of proline in the plant (Marschner, 1995). Clinoptilolite zeolite is a suitable adsorbent of ammonium cation and reduces the leaching of nitrogen from the root (Polat et al. 2004). In some studies, the use of zeolite led to an increase in the proline content (Mohammadzadeh and Pirzad, 2014). However, Ramjerd et al. (2015) showed in an experiment that zeolite led to the reduction of proline.

In the present experiment, fifty percent of the treatments (ten treatments marked with the letter C in Figure 5) did not have a

significant difference with the control treatment. In applying high levels of zeolite fertilizer, a large difference with the control treatment was observed in the production of proline. In the present research, compost fertilizer was less effective than zeolite in increasing the proline content. Ahmadian et al. (2011) stated that the increase of soil nitrogen in manure treatments increased proline in *Matricaria chamomilla*. It can be said that applying high levels of fertilizer can increase nitrogen to the point of toxicity in the plant which reacts by producing proline.

Root dry weight

In general, in our study, both types of fertilizers had a positive effect on root dry weight. Gruener et al. (2007) stated that treatments containing zeolite with cation exchanges and the substitution of ammonium and potassium by calcium, in addition to supplying potassium and ammonium to plants, dissolve phosphorus in the soil solution and thus provide the plants with their needed nutrients. Probably for the same reason, the plant roots absorb more food in the presence of zeolite and thus have more weight than the control treatment. The results of this experiment are in accordance with the results of Ahmadi Azar et al. (2015) but are incompatible with those of Gholizadeh (2006).

Compost, due to having large amounts of organic matter, improves the physical and chemical properties of soil and thus makes it easier for plants to access nutrients. Sharifi et al. (2010) stated that manure improved soil conditions and increased root dry weight.

Shoot dry weight

The use of zeolite in manure supplies the plant with nitrogen and increases the performance and the dry weight of the shoots by keeping nitrogen in the fertilizer mass. Khan et al. (2009) stated that compared with the control treatment, zeolite treatments increased shoot dry weight. The positive effects of manure, especially increasing soil moisture retention capacity and improving the nutritional conditions of the plant increase the dry

weight of the plant (Malakouti and Sepehr, 2004). In another experiment by Sharifi et al. (2010), an increased leaf dry weight was reported as a result of applying manure.

Finally, it can be said that the interaction effect of the applied fertilizer levels improved the plant performance. However, it should be noted that applying high levels of fertilizer can cause toxicity in plants due to increasing nitrogen through the production of proline. It is suggested that the long-term interaction effect of these fertilizer levels on rangelands be examined.

Conclusion

The results of this study showed that the use of zeolite clinoptilolite with compost manure had a significant effect on all the

measured traits compared to the control treatment. The interaction of these fertilizers not only prevented the loss of the nitrogen compounds of soil and preserved this valuable nutrient but also increased the quantitative and qualitative performance of Vetiver grass. Since both of these fertilizers are natural fertilizers, less chemical fertilizers will be used preventing damage to the environment. The use of this kind of soil amendment can be recommended for the optimal growth of Vetiver grass in degraded rangelands. Nevertheless, to obtain more comprehensive results and to study the long-term effect of fertilizers on soil and plants, similar tests should be conducted in the rangelands under different environmental conditions.

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