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Determination of Allometric Relationships in two Basil Ecotypes as Affected by Organic Fertilizers

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ABSTRACT

The geometric shape of a plant and its components depend on the allocation of absorbed nutrients and substances to a growing organ. In turn, the overall outcome is influenced by genetics and local environmental conditions. The relationship between different components of an organ can be shown with the help of allometry. To determine the effects of organic fertilizers on morphological traits and yield of basil, this research was conducted as a factorial experiment based on a Completely Randomized Design (RCD) with three replication -s under controlled conditions. Experimental factors were basil ecotypes (green and purple) and organic fertilizers (cow manure, vermicompost, sulfur granular compost, and control). Results indicated that the highest values of plant height, fresh and dry weight of lateral branches, leaf count, leaf area, leaf fresh and dry weight, and plant biomass were found in green basil when vermicompost was applied. In contrast, the lowest mean values of these parameters were observed in the control. The application of vermicompost, cow manure, and sulfur granular compost caused increases of 30, 29, and 28% in the plant biomass of purple basil. Meanwhile, they caused increases of 61, 49, and 28% in the plant biomass of green basil, respectively. Also, the fitted experimental allometric relationship indicated a significant correlation between average leaf count and average leaf area (r^2=0.95). Allometric relations-hips between root and shoot dry weight also indicated a more symmetric growth of these two parameters in green basil, compared to purple basil. Therefore, the analysis of growth relationships between plant components and production could be evaluated based on morphological components (i.e. leaf count, leaf area, and root dry weigh t as well as shoot dry weight), and allometric equations.

Introduction

Basil (*Ocimum basilicum* L.) is an annual herb that belongs to the Lamiaceae family. The *Ocimum* genus has 30 species. *Ocimum basilicum* is the most prevalent species in terms of commercial importance in the Americas, Spain, Pakistan, Indonesia, and almost all warm and temperate areas (Marotti et al., 1996). Basil is used for its leaves, flower heads, and seeds. Fresh and dry leaves of this plant are widely used in food industries and its essential oil is applied in the cosmetics, sanitary, and perfumery industries (Beatovic et al., 2015).

In the 70s, the application of chemical fertilizers

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resulted in higher crop yield, but greenhouse gas emissions using nitrogen fertilizers and N2O from these fertilizers disturbed ecosystem sustainability (Abdalla et al., 2016). Also, in some regions, the deficiency of soil organic matter undermined the sustainability of agroecosystems. Unsuitable agronomic management led to lower soil fertility, increased environmental problems, and groundwater pollution. On the other hand, because of a decrease in agricultural product quality, vitamin content and other plant-useful compounds have decreased in response to the irresponsible use of chemical fertilizer applications (Atiyeh et al., 2000).

The application of manure is known to have a special place in agriculture. Organic matter is one of the main soil components which improves physical, chemical, and biological characteristics of soils as well as the processes that occur in them. It provides nutrients needed for plant growth (Madrid et al., 2007). Organic fertilizers have many useful microorganisms. Their metabolic products could convert the inaccessible form of nutrients into accessible forms during biological processes, thereby improving soil fertility, seed germination, root system development, plant biomass, and, finally, an increase in crop qualitative and quantitative yield (Rajendran and Devaraj, 2004). As an organic fertilizer, compost is produced from urban, agricultural, or livestock wastes through fermentation. It has no harmful effect on agricultural soils and, in fact, improves soil physical and chemical criteria. Its beneficial nutrients are released gradually and continuously in the soil and are made available to plants (Greenberg et al., 2017). Vermicompost is also another organic fertilizer that is the product of organic matter digestion by earthworms (*Eisenia foetida*). Vermicompost contains hormones and enzymes which help the natural recycling of materials and prepare nutrients for plant absorption (Senesi, 1989).

Increasing soil organic compounds and supplying essential nutrients for plant growth such as potassium followed by yield improvement, as a result of vermicompost amendments to soils, have been reported by Sinha et al. (2009). Reports are also available on the effective role of vermicompost in converting NH_4^+ to NO_3^- (Gutiérrez-Miceli et al., 2007). Thus, it could be concluded that vermicompost can control soil pH and inhibit the rise of pH through NH_4^+ reduction. Darzi and Hadi (2012) reported that the seed yield of anise (*Pimpinella anisum* L.) was increased significantly by the application of vermicompost treatments. Fe deficiency with symptoms of chlorosis in young leaves is one of the important nutritional failures in calcareous soil, which severely affects crop yield and quality, and, finally, could lead to crop loss (Secco et al., 2017). Kalbasl et al. (1986) reported that using sulfur granular compost could assist with the local acidification of soil and increase solubility and absorption of micronutrients (e.g. Fe and Zn). It has been indicated that the simultaneous application of sulfur and organic fertilizers has a more pronounced effect on crop yield and enhanced absorbency of soil nutrients, compared to the use of sulfur per se (Cifuentes and Lindemann, 1993). An increased plant height followed by an increase in fresh and dry matter yield was observed in basil when organic fertilizers were used (Tahami Zarandi et al., 2010).

Despite that plant growth can be affected by planting beds, the geometric shape of the plant and its components is always the same. Allometry is defined as the relationships between the growth rate of individual components of an organ or an organism. In other words, allometry aims to assess the allocation of assimilates to a growing organ which, in turn, is dependent on genetic and environmental conditions (Robinson et al., 2010). Leaf area is one of the most important variables, used in studying growth parameters, vital processes, and energy balance in the plant (Ma et al., 1992). Therefore, leaf area measurement and finding the allometric relationships between these parameters and the other plant vegetative components could assist researchers in reaching a better understanding of the effects of environmental factors on plant growth (Tobin et al., 2006). Accordingly, this experiment was conducted to assess morphological characteristics and allometric relationships in basil plants by the application of organic fertilizer.

Material and Methods

This research was conducted as a factorial experiment, based on CRD, with three replications at the Faculty of Agriculture, Ferdowsi University of Mashhad (spring 2019). The first factor was two basil ecotypes (green and purple) and the second was four organic fertilizers (cow manure, vermicompost, sulfur granular compost, and control). Seeds were planted in each 10 kg pot, filled with equal proportions of sand, silt, and clay. The nutrient contents of soil and fertilizers were determined (Table 1). The amount of fertilizer needed to add to the soil was calculated based on soil analysis. Thus, 150, 60, and 80 g of calibrated (2 mm diameter) cow manure, vermicompost, and sulfur granular compost were obtained by sieving. These were added to each pot based on the treatments. Ten seeds were planted at equal distances in each pot and were thinned to five plants per pot. The amount of irrigation water was considered the same for all treatments. Relative contents of chlorophyll (SPAD readings) were assessed by a chlorophyll meter (Mintola Reading SPAD, Japan) at 50% flowering. Plant height, branch count and length, leaf count, and leaf area (using AT; Delta-T Devices Ltd, leaf area meter) were considered in addition to the length and number of inflorescence, main stem, lateral branches, and leaf fresh weight. Root and shoot dry weights were determined at the end of the growing season.

Table 1. Physical and chemical properties of soil, cow manure, vermicompost, and sulfur granular compost

Sample	pH	EC (dS m^{-1})	Nitrogen	Phosphorous	Potassium
Soil	7.3	2.2	0.0014%	0.00123%	0.0376%
Cow manure	6.8	5.6	1.4%	1.3%	1.6%
Vermicompost	7.6	3.5	1.5%	1.3%	1.7%
Sulfur granular compost	6.5	4.5	0.7%	0.4%	0.8%

The allometric power function between leaf area (Y) and leaf count (X) was determined by eq. 1:

$$Y=aXb$$

Where a and b are intercept and line-slope, respectively. The coefficient of variations (CV) was used for assessing the accuracy of the equation (Tobin et al., 2006). Allometric relations between root and shoot in young herbal plants were described using eq. 2. Eq. 2:

$$R=\beta s\alpha$$

Where R, s, β, and α are the dry weight of root and shoot, allometric coefficient, and scaling exponent, respectively.

Despite the precision in setting the number of plants, it was less than the desired density in some pots, and, thus, covariance analysis was done based on the number of plants, before the analysis of variance. The analysis of variance was done by MINITAB ver. 13 and graphs were drawn by Microsoft Excel. Mean values were compared using the LSD test ($P \leq 0.05$).

Results

The results showed that the amount of chlorophyll content (SPAD) changed differently in each of the basil ecotypes, although the trends of changes were almost similar in them. Thus, organic vermicompost, cow manure, and sulfur-granulated compost in green basil led to SPAD values of 48.1, 41.6, and 35.2, respectively. In purple basil, the SPAD value became 43.9, 42.6, and 33.8, respectively. The SPAD value in the control treatment in both basils was 39 (Fig. 1).

The highest plant height was found in green basil by the effect of vermicompost treatment, whereas the lowest belonged to the purple ecotype with no fertilizer (Table 2).

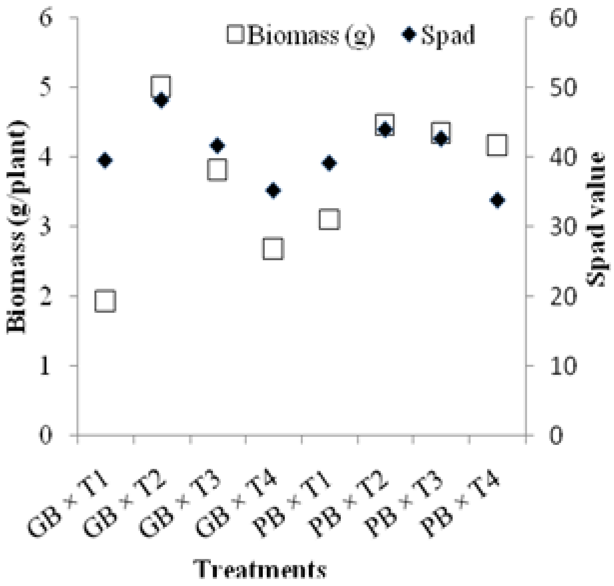

Fig. 1. Interaction of organic fertilizer × basil ecotypes on biomass and SPAD values. GB: green basil, PB: purple basil, T1: control, T2: vermicompost, T3: cow manure, T4: sulfur granular compost

Table 2. Mean values of plant height, fresh and dry weights of stems, branch count and length, fresh and dry weights of branches, leaf fresh and dry weights, leaf count and surface area, biomass, root dry weight, and SPAD value of the two basil ecotypes and manure treatments

	Green Basil				Purple Basil			
	T1	T2	T3	T4	T1	T2	T3	T4
SPAD	39.5^{c}	48.1^{a}	41.6^{b}	35.2^{d}	39.1^{c}	43.9^{b}	42.6^{b}	33.8^{d}
Plant height (cm)	38.7^{bc}	47.4^{a}	42.9^{ab}	37.5^{bc}	32.9^{c}	39.7^{a-c}	37.3^{bc}	44.7^{ab}
Number of branches per plant	0.76^{f}	7.80^{b}	8.15^{b}	2.36^{e}	7.23^{bc}	10.30^{a}	6.29^{c}	5.10^{d}
Length of branch (cm)	13.4^{f}	103.1^{c}	83.8^{d}	26.3^{e}	97.4^{c}	115.9^{a}	109.5^{b}	102.3^{c}
Leaf number per plant	25.9^{e}	124.2^{a}	53.2^{d}	116.3^{ab}	86.4^{c}	107.5^{b}	116.5^{ab}	115.9^{ab}
Leaf area (cm^2 $plant^{-1}$)	87.1^{e}	424.7^{a}	126.3^{d}	358.0^{a}	211.2^{c}	308.6^{bc}	381.1^{ab}	401.0^{ab}
Dry weight of main stem (g $plant^{-1}$)	0.754^{d}	1.310^{a}	0.960^{c}	0.798^{d}	0.783^{d}	1.120^{b}	0.885^{c}	0.864^{c}
Fresh weight of main stem (g $plant^{-1}$)	2.23^{d}	5.62^{a}	3.14^{c}	2.65^{d}	2.36^{d}	4.71^{b}	3.58^{c}	3.46^{c}
Dry weight of lateral branches (g $plant^{-1}$)	0.213^{f}	1.490^{a}	0.418^{e}	1.180^{b}	0.767^{d}	1.250^{ab}	1.130^{b}	0.907^{c}
Leaf dry weight (g $plant^{-1}$)	0.83^{e}	2.78^{a}	1.03^{d}	1.76^{cd}	1.30^{d}	2.30^{ab}	2.25^{bc}	1.95^{bc}
Biomass (g $plant^{-1}$)	1.93^{e}	5.01^{a}	3.81^{c}	2.68^{d}	3.10^{bc}	4.46^{b}	4.34^{b}	4.17^{bc}
Root dry weight (g $plant^{-1}$)	1.06^{f}	2.80^{a}	1.94^{cd}	1.50^{e}	1.64^{de}	2.34^{b}	2.24^{b}	2.03^{c}

Mean values with the same letters in each row are not significantly different ($P \leq 0.05$) based on Duncan's multiple range test.

T1: Control; T2: Vermicompost; T3: Cow manure; T4: Sulfur granular compost.

The highest and lowest number and length of lateral branches were obtained in purple basil by applying vermicompost, and in green basil with no fertilizer, respectively. The application of vermicompost caused 24 and 11% increases in the number and length of lateral branches in purple and green basil, respectively, compared to the control (Table 2). The highest leaf count belonged to the vermicompost treatment in green basil. There was no significant difference between the effects of cow manure and sulfur granular in purple and green basil. The lowest value of this parameter was found in green basil with no fertilizer, which was 70% lower compared to purple basil in this fertilizer treatment (Table 2). The application of vermicompost caused an increase of 79 and 31% in leaf area in green and purple basil, respectively, compared to the control. Leaf area was increased by about 27% higher in green basil, compared with the purple basil, in the vermicompost treatment (Table 2). The highest and lowest dry weight of the main stem was found in green basil, as a result of applying vermicompost, and in the control, respectively. In this trait, no significant difference was observed between the two ecotypes when cow manure was used. The lowest dry weight of lateral branches per plant was observed in the green and purple basil under the control treatment, respectively. This parameter was increased in the green and purple ecotypes by 86 and 39% via the application of vermicompost, respectively (Table 2). The highest dry leaf weight was obtained in the green basil and the application of vermicompost, which became 30% higher, compared to the purple, and 70% higher compared to the control. The lowest dry leaf weight was found in green basil with no fertilizer. When cow manure and sulfur granular compost were applied, the dry leaf weight of purple basil increased by 42 and 23% in comparison with the control, respectively. The highest and lowest biomass values were found in green basil, as a result of the vermicompost treatment and in the control, respectively. The application of vermicompost, cow manure, and sulfur granular compost increased the biomass up to 30, 29, and 26% in purple basil, and up to 61, 49, and 28% in green basil (Table 2). These results showed that the response of the green basil ecotype was about twice as much as the response observed in the purple basil, under the vermicompost treatment. The effects of cow manure on basil biomass were more relevant in the green ecotype, compared to the purple basil, but the effect of sulfur granular compost was almost the same on both (Table 2).

In this study, there was an exponential relationship (y=0.1x1.734) between leaf count and leaf area (Fig. 2). The coefficient of variation (CV) for leaf area per plant, based on leaf count per the main stem, was 10.7% with r2=0.95, showing a high accuracy of the fitted curve. Allometric relationships between root and shoot dry weight showed a significantly higher α value in the purple ecotype, compared to the green basil. In green basil, since α=1.021, a balanced growth ratio was indicated between root and shoot. The allometric coefficient was significantly different between the two ecotypes (Table 3).

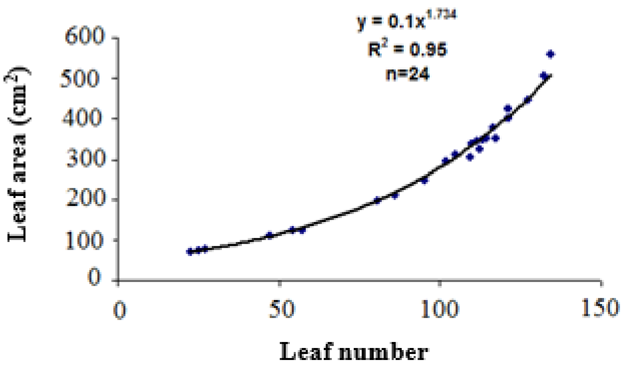

Fig. 2. Exponential regression fitted between average leaf count and average leaf area in the two basil ecotypes (green and purple)

Table 3. Allometric coefficients in the two basil ecotypes

Ecotype	Allometric equation	β	α	R^2
Green Basil	$R=0.528S^{1.021}$	0.528	1.021	0.99
Purple Basil	$R=0.556S^{0.943}$	0.556	0.943	0.94

Discussion

Many methods can be used for modifying the physical and chemical structures of soils to increase soil fertility and plant growth. One of these methods is adding organic fertilizers (Arcon et al., 2005). Among the various sources of organic fertilizers, vermicompost, cow manure, and granulated sulfur-containing compost fertilizer are regarded as potential soil amendments. They provide micro and macro elements for the rhizosphere zone. Organic materials with a cation exchange capacity of up to 300 cmol kg^{-1} and a specific surface area of up to 900 square m kg^{-1} have a high potential for retaining and releasing nutrients. They play a major role in the maintenance of soil acidity (Wang and Yang, 2002). The results of the present study showed that in both basil ecotypes, the amount of biomass increased with the application of organic fertilizers, compared with the control. The organic fertilizer had a greater effect on the purple basil ecotype than on the green basil, although, among all treatments, the vermicompost treatment had a significant effect on biomass production in green basil. The results indicated that the application of vermicompost in fennel (*Foeniculum vulgare*) (Darzi and Hadi, 2012) and dill (*Anethum graveolens*) (Shahmohammadi et al., 2014) increased plant yield. The application of vermicompost also led to a high yield, compared to the effect of cow manure in basil (Tahami Zarandi et al., 2010). In a study by (Alidadi et al., 2013), the application of 4 $tons.ha^{-1}$ sulfur granular compost increased saffron (*Crocus sativus*) yield, compared to the effect of vermicompost, whereas saffron yield decreased substantially as the amount of fertilizer increased to 8 $tons.ha^{-1}$. The compost fertilizer increased soil microbial activity and when sulfur was added to these fertilizers, the activity of sulfur bacterial oxidation led to a decrease in pH, organic acid protection, and soil organic solubility of Fe matter mineralization. A decrease in soil pH reportedly increased the solubility of Fe and reduced the solubility of K and Ca (Sharpley, 1991), which seems to be a suitable condition for basil. Reports have indicated that vermicompost leads to a more porous soil texture and a lower soil bulk density, thereby increasing soil water capacity (Han et al., 2016). On the other hand, changing the amounts of organic matter residue increases the rate of decomposition, which has positive effects on the fixation and conversion of elements needed by plants. Ultimately, this provides more suitable conditions for plant growth and increases the biomass of medicinal plants (Egamberdieva et al., 2015). Plant height is increased by organic fertilizers (Tahami Zarandi et al., 2010). It seems that organic fertilizers, with increased root absorption surface for each unit volume of soil, had direct effects on photosynthesis and other physiologic processes, leading to more plant growth and higher plant height. In another report, Zinati et al. (2004) suggested that animal manure reduced acidity but increased electrical conductivity, organic carbon, total nitrogen, phosphorus, potassium, iron, and zinc availability in the soil. The findings of this study showed that the application of organic fertilizers in both ecotypes increased the dry weight of the leaves and the dry weight of the lateral branches, compared to the control, and that the greatest increase was achieved in response to the vermicompost treatment in both ecotypes. Tahami Zarandi et al. (2010) reported that the highest dry leaf weight of basil occurred because of vermicompost and that there was no significant difference between cow manure and hen manure. It seems that the presence of a large microbial population in vermicompost assists in a considerable buildup of microbial populations and microbial activity in the soil, which influenced plant growth indirectly (Arancon et al., 2005).

In our study, the organic fertilizer increased the number of branches per plant in both basil ecotypes. Atiyeh et al. (2000) reported a significant increase in the number of the main stems and lateral branches of potatoes in vermicompost treatments. Rezaee-Moadab et al. (2012) indicated that the application of vermicompost caused an increase of 17.3% in lateral branches of basil (*Ocimum basilicum* L.), 21% higher than that obtained by the application of chemical fertilizer and the control treatment. In the current research, the highest dry weight of the main stem was found in green basil in vermicompost treatments. This is consistent with previous results by Atiyeh et al. (2000), which indicated that the application of vermicompost and compost increased the dry stem weight of basil. The application of vermicompost resulted in an increase of 16% in the root dry weight of green basil, compared to that of purple basil. In a study by Weisany et al. (2012), a total of 90 and 111 leaves (37 and 49% higher than the control) were obtained as a result of using biological fertilizers nitroxin and nitroxin + phosphate solubilization bacteria, respectively. Also, the leaf count per plant in this research was 83, as a result of applying chemical fertilizers. The presence of more pores and proper ventilation, because of organic fertilizers, especially vermicompost, led to a greater activity of microorganisms in the soil, thereby providing nitrification conditions and improving nitrate absorption by plants. The absorbed nitrogen is usually assimilated into amino acids, proteins, and nucleic acid structures, thereby increasing cytokinin biosynthesis indirectly, and, therefore, increasing the production of new tissues and plant growth.

There is an allometric relationship between leaf area and plant vegetative components, e.g. leaf count, plant height, shoot dry weight, and leaf dry weight (Niklas, 1994). In this study, the coefficient of variation was r^2=0.95 which shows the high accuracy of the fitted curve. In a study by Rahimi Karizaki et al. (2006), different fittings of leaf area, based on vegetative criteria, were reported between 0.8-0.97, and the coefficient of variation was in the range of 5.4-12.5%. Robinson et al. (2010) reported an exponential relationship (R=βsα) between shoot and root dry weight in two herbal species of *Plantago lanceolata* and *Dactylis glomerata*. Allometric models could be useful in estimating plant growth, although root and shoot growth in each species is regulated genetically and environmentally, in addition to the effects of physiological and ecological parameters (Enquist and Niklas, 2002).

Conclusion

Generally, the results of the present study indicated the positive effects of organic fertilizers on plant growth. By applying the vermicompost treatment, plant biomass increased in both ecotypes, compared to the control and other fertilizer treatment groups. The fitted equation between leaf count and leaf area indicated an exponential relationship ($y=0.1x^{1.734}$) by which leaf area could be estimated in correlation with leaf count. In this study, the highest leaf count and the largest leaf area occurred in response to the vermicompost treatment. On the other hand, studying the allometric relationship between root and shoot dry matter indicated a symmetric growth of roots and shoots in green basil, whereas it showed more root growth in purple basil. Evidently, the application of vermicompost increased biomass production in green basil by enhancing morphological criteria such as leaf count and leaf area.

Conflict of interest

The authors indicate no conflict of interest for this work.

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