Comparative analysis of growth parameters of protected organic leafy vegetables

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Abstract: This study was focused to evaluate effect of organics on the growth parameters of three different green leafy vegetables (Indian spinach, Fenugreek and Amaranthus) under protected systems. Split-split plot design was employed to study the effects of the system (S), organic treatment (NM), crop types (C) and their interaction on the growth parameters. Net assimilation rate (NAR) and total dry weight (DW) were significantly higher (p≤0.05) in the polyhouse compared to shadenet for all three crops. System: treatment interaction was also significant at the level of p≤0.05. Crops treated with vermicompost performed significantly (p≤0.05) better in both the systems at all 3 growth stages. Crops maturity advanced by 10 days in the polyhouse compared to the shadenet. Comparatively all 3 crops performed better at the level of p≤0.05 in terms of their growth parameters in the polyhouse than in the shadenet.

Keywords: Growth parameters - Polyhouse - Shadenet - Split-split plot - Leafy Vegetables - Organics.

INTRODUCTION

In recent decades, the use of polyhouse for growing vegetable crops has increased worldwide. Partially or fully protected cultivation under polyhouses and shadenets are preferred by urban growers with small land holdings to increase the crop production (Sabir & Singh 2013) by controlling disease and pest incidences (Nordey et al. 2017). Vegetables are grown all round-year irrespective of climatic condition for better price for the growers (Singh & Sirohi 2004) under protected cultivation. Even at higher altitudes, vegetables in protected cultivation has proven successful in providing regular food supply (Mishra et al. 2010) and improved food quality thus benefiting farmers with small landholdings (Chang et al. 2013) in developing countries. Another reason for considering organic treatment to protected leafy vegetables as such studies with respected organic production is limited, although crops in response to organic production physiologically accumulate organic molecules (antioxidants) which might have potential health benefits (Orsini et al. 2016). Influence of greenhouse on evapotranspiration has been reported minimum in shallow hall including in polyhouse and shadenet (Santosh et al. 2017). Growing in high-tech greenhouses, avoiding strong dependence on the outdoor climate which helps in supplying high-quality vegetables all year round (Qaryouti 2013). Temperature and relative humidity in shade net are slightly higher than the outside conditions; however outside weather condition still have a direct influence on it. Shadenet houses are suitable for warm climatic condition for growing vegetables during the summer months. Such influence on physiological processes in tomato in protected systems with organic treatments have be found strongly correlated with the dry biomass produced (Acatrinei 2010). Protected cultivation is found to be suitable technology for seasonal and off-seasonal vegetable cultivation, particularly at high-altitude region to increase the yield significantly as compared to that in open field condition (Negi et al. 2013). Therefore growth parameters analysis of three protected organic green leafy vegetables was carried as to evaluate which protected system is suitable for organic production.
MATERIALS AND METHODS

Crops

Green leafy vegetables are the main source of minerals (including iron, calcium, potassium, and magnesium and vitamins, including vitamins K, C, E, and many of the B vitamins (Jiménez-Aguilar & Grusak 2017, Singh et al. 2018). Indian spinach (Beta vulgaris var bengalensis Roxb.) with succulent leaves is one of the most popular leafy vegetables of the tropical and subtropical region and is grown widely in India (Padmanabha et al. 2008). The other two selected vegetables were Fenugreek and Amaranthus. Trigonella foenum-graecum L. (Fenugreek) is native to Asia and southern Europe known for culinary and medicinal uses in the history of old civilizations (Khan et al. 2014). Amaranthus sp. (Amaranth) is an annual or short-lived perennial plant used as leafy vegetables, grain, ornamental, medicinal and forage crop in many countries including India (Haupl & Jain 1977, George et al. 2014). Since the above short duration leafy vegetables are predominantly and preferably grown by the farmers with small landholdings, choosing these vegetables were appropriate for the experiment that aimed to compare growth parameters of leafy vegetables under two types of the protected system. Local organic seeds of Indian Spinach (C1), Fenugreek (C2) and Amaranthus (C3) were collected from Organic farmer of Dharwad, North Karnataka, India.

Field site and growing condition

This experiment was conducted in two different partially protected systems (polyhouse and green colored shadenet (40% shade factor), during March and April, 2018 at Bioresource Farm of Institute of Organic Farming, University of Agricultural Sciences, Dharwad, Karnataka, India. The organic field site (Plot No: C 52-A; Coordinates: 15°29′50.695″N; 74°58′40.966″E, with elevation 768.0 m) was used for cultivation under the constant care of a gardener. A sprinkler-can was used for normal irrigation. The experiment was laid out on red soil and medium black loam soil in Polyhouse (S1) and Shadenet (S2) respectively. In Dharwad, bulk density, percent pore space, moisture content, organic matter content, Electrical Conductivity (EC) and available K are considered higher in black soil compared to red soil with soil pH ranging from neutral to alkaline and acidic to neutral (Bidari et al. 2008).

Weather condition

Weather data was collected from the Department of Agricultural Meteorology, University of Agriculture Sciences, Dharwad, Karnataka, India. The average recorded temperatures were 35.2°C and 39.5°C in S1 and 36.2°C and 38.5 in S2 for the month of March and April respectively, whereas, the maximum average temperature was 34.9°C (March) and 36.2°C (April) and minimum was 19.3°C (March) and 21.1°C (April) at University of Agriculture Sciences, Dharwad. The average recorded relative humidity (%) were 43% and 41% for the month of March and 34.5% and 35.3% for the month of April in S1 and S2 respectively. Outside minimum and maximum average relative humidity recorded 65.3% and 79.6% for the month of March and 27.8% and 36.7% for April. Temperature in S1 was comparatively higher than S2 and outside temperature while relative humidity was opposite during the study period.

Experimental design

| Crop        | N (kg ha⁻¹) | P (kg ha⁻¹) | K (kg ha⁻¹) |
|-------------|-------------|-------------|-------------|
| Spinach (C1)| 150.00      | 100.00      | 100.00      |
| Fenugreek(C2)| 100.00     | 50.00       | 0.00        |
| Amaranthus(C3)| 100.00    | 50.00       | 50.00       |

Split-split plot design was used. S1 and S2 as the two main plots, where type of crops (C1, C2 and C3) as the split-plot and organic treatments: Control (NM1), Vermicompost (NM2), Vermicompost (VC)+Neem cake (NM3), VC+Neem cake (NC)+Rock phosphate (NM4) and compost (NM5) product of Institute of Organic Farming, UAS, Dharwad) as the split-split plot. Each plot length of 18x1 m in 3 replications were equally divided into 5 sub-sub plots. There were two systems, three crops and five treatments resulting to 30 observations per replicate (2x3x5). The experiment was performed in three replicates over 40 days. Organics were applied based on the recommended (Sumpena & Hidayat 1999, Islam et al. 2011, Qaryouti 2013) doses (Table 1) for each type of crop. Seeds were shown in four rows with outer row spacing of 11.5 cm and inter-row spacing of 22.5 cm.

Soil and seed bed preparation

Soil beds were prepared using simple tools like spade and handheld harrow. Coarse sand and stones were separated and removed from the beds. Organics were applied 7 days before sowing of seeds. All three crops
were sown by hand dibbling method on 10 March. Previous crop history: In S1 trial plot tomato and cucumber were grown. There was no immediate record of the previous crop grown in S2 trial plot.

Data gathering

Growth parameters (Leaf Area Ratio, Leaf Weight Ratio, Specific Leaf Weight, Specific Leaf Area, Net Assimilation Rate and Dry Weight) were recorded at 20 DAS (days after sowing), 30 DAS and 40 DAS (harvest) for 3 growth stages (GSs). Crops were sown on 10 March and harvested on 20 April 2018.

RESULTS

All analysis was performed in triplicates (n= 3) corresponded to both the protected systems (S1 and S2). Two-way analysis of variance (ANOVA) with mean separation by Tukey’s Least Significant Difference (LSD) at α = 0.05 was used to determine significant differences between systems (S1/2), treatments (NM−5), and their interactions (S1/2 × NM−5). Mean among S1/2, NM−5 and their S1/2 × NM−5 were compared using LSD at p ≤0.05. Data analysis was done using R (R Core Team 2018) with agricolae package (de Mendiburu 2017). Results are given crop-wise comparing growth parameters between two systems for 3 GSs. Two-way analysis of variance (ANOVA) carried out with mean separation by Tukey’s Least Significant Difference (LSD) at p ≤0.05 level of significance.

Crops

The effect of systems on the growth parameter like LAR, LWR and SLA of the green leafy vegetables were statistically significant at all three stages as shown in tables 2 & 3. Further, LAR, LWR and SLA were significantly at the level of p ≤0.05 for different crops at all stages. The effect of organics on the growth parameter for three different leafy vegetables at 20DAS were not significantly different from control (NM−5) except for treatment (NM5) which has significant difference in the mean compared to control.

Table 2. Effects of protected systems and organic nutrients on LAR, LWR and SLA at 20, 30 and 40 DAS (harvest) of green leafy vegetables.

| Mean values | LAR (cm² g⁻¹) | LWR (g g⁻¹) | SLA (cm² g⁻¹) |
|-------------|---------------|-------------|---------------|
| Growth Stages | 20DAS 30DAS Harvest | 20DAS 30DAS Harvest | 20DAS 30DAS Harvest |
| Protected Systems (S) | | | |
| Polyhouse (S1) | 15.63b 26.84b 37.83b | 0.48b 0.17b 0.15b | 72.54a 53.95a 37.83b |
| Shadenet (S2) | 16.89a 31.41a 61.23a | 0.52a 0.19a 0.16a | 76.38a 55.36a 61.23a |
| LSD at 5% | 1.22 3.64 4.85 | 0.03 0.01 0.01 | 4.62 5.48 4.85 |
| Crops (C) | | | |
| Spinach (C1) | 20.32a 45.33a 61.05a | 0.56a 0.20a 0.19a | 84.02a 74.26a 61.05a |
| Fenugreek (C2) | 15.41b 26.92b 44.82b | 0.51b 0.20b 0.18b | 70.16b 46.02b 44.82b |
| Amaranthus (C3) | 13.06c 15.12c 42.73c | 0.44c 0.13b 0.11b | 69.20b 43.69b 42.73b |
| LSD at 5% | 1.12 4.38 6.64 | 0.04 0.02 0.01 | 5.93 7.59 6.64 |
| Nutrient management (NM) | | | |
| Control (NM−5) | 16.57b 27.83ab 49.19ab | 0.52ab 0.18a 0.18b | 73.93ab 51.33a 49.19ab |
| VC (NM−4) | 14.68b 25.80b 48.64b | 0.49bc 0.15b 0.13c | 70.99b 58.17a 48.64b |
| VC+NC (NM−3) | 15.00b 33.04a 48.20b | 0.50bc 0.18a 0.16ab | 67.94b 58.26a 48.20b |
| VC+NC+RP (NM−2) | 5.93b 32.98a 45.94b | 0.46b 0.19a 0.17b | 77.24b 54.83a 45.94b |
| Compost (NM−1) | 19.14a 25.96b 55.70a | 0.54a 0.18a 0.17a | 82.21a 50.68a 55.70a |
| LSD at 5% | 1.95 5.77 8.16 | 0.04 0.07 0.01 | 10.05 10.92 8.16 |

Note: abcValues (column means) with the different small letter in a column in DAS are significantly different according to the LSD test at α = 0.05; NM−5Vermicompost (VC), Neem Cake (NC), Rock Phosphate (RP), Compost, Days after sowing (DAS) and Growth Stages (GSs).

Spinach (C1):

Growth parameters of C1 were significantly higher at the level of p<0.001 in polyhouse (S1) than in shadenet (S2). C1 performed much better in terms of DW in S1 compared to S2. Growth parameters also steadily increased till 40 DAS (harvest) in both systems. C1 attained early crop maturity in S1 than in S2 at by 10 DAS.

Fenugreek (C2):

C2 also performed better in S1 compared to S2. Early maturity of C2 was also observed in S1 compared to S2. DW was also significantly higher at the level of p ≤0.05 in S1.

Amaranthus (C3):

Similarly, growth parameters of C3 are significantly higher in polyhouse (S1) than in shadenet (S2). DW was significantly higher in S1 compared to S2. C3 attained early maturity in S1 than in S2 at least by 10 DAS.

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Modern tools for classical plant growth analysis have been proposed by many authors. The procedure and analysis in this study followed classical tools which are covered under the method section. Graphical display of pair-wise comparisons from Tukey’s HSD for the growth analysis. Any confidence intervals that do not contain

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provide evidence of a difference in the groups. Plant samples were collected at 20, 30 (DAS) and harvest for growth analysis. To measure and analyse the effects of protected systems and organic nutrients on LAR, LWR, SLW, SLA, NAR and DW (gm) of green leafy vegetables for 3 GSs. Tukey’s Honest Significant Difference (Tukey’s HSD) method to make all the pair-wise comparisons was used to compare all the means of LAR, LWR, SLW, SLA, NAR and DW. In general LSD test for growth parameters were significant over growth period and between interactions (Table 2 & 3). Total dry matter production was comparatively higher (p ≤0.05) in NM3 followed by NM2 and NM5. Total DW was significantly low for NM4 followed by NM1. The yield of a field crop is the weight per unit area of harvested produce; thus it is logical to base an analysis on weight changes (Wolf & Carson 1973). Plant growth analysis is an explanatory, holistic and integrative approach to interpreting plant form and function (Hunt et al. 2002).

Three sample plants at 3 GSs (20, 30 (DAS) and harvest) representing every sub-sub plot and treatment were collected and brought to the lab for further growth analysis. Stems, leaves and roots of every plant sample were separated and left for shade drying for 5 days. Overnight oven drying was done at 45°C. Growth parameters were measured as per (LAR (cm$^2$ g$^{-1}$), Radford (1967)), (LWR (g g$^{-1}$) and SLA (cm$^2$ g$^{-1}$), Kvet & Marshall (1971)), (SLW (g cm$^{-2}$), Pearce et al. (1969)), (NAR (g g$^{-1}$ t$^{-1}$), Williams (1946)) and total DW (gm) at 20, 30 (DAS) and harvest estimated after oven drying.

$LAR \ (cm^2 \ g^{-1})$

At 20 DAS Leaf Area Ratio was significant for treatment and crops at the level of *p<0.05 and ***p<0.001 respectively. LAR was higher in S2 compared to S1. Later at the harvest stage LAR was significant for systems, crops and their interaction at the level of ***p<0.001 (Fig. 1). Adjusted R$^2$ for LAR recorded was low at 0.319, 0.604 and 0.486 at 20, 30 (DAS) and harvest respectively. Adjusted R$^2$ was 0.614 for LAR mean of 3 GSs.

$LWR \ (g \ g^{-1})$

Leaf Weight Ratio was another growth parameters measured for the analysis which was significant for treatments, system and crops at the level of ***p<0.001 for all 3 GSs (Fig. 2). LWR was higher in S2 compared to S1. However interaction between (S1/2 : NM1−5) was not significant (Table 3). Adjusted R$^2$ for LWR was 0.884, 0.526 and 0.671 at 20, 30 (DAS) and harvest respectively. Adjusted R$^2$ was 0.717 for LWR mean of 3 GSs. LWR was significant (P ≤0.05) for 3 GSs.

$SLA \ (cm^2 \ g^{-1})$

Specific Leaf Weight was significant with system and crops at the level of ***p<0.001 for 20 and 30 DAS. (S1/2  : NM1−5) the interaction was significant only at harvest (Fig. 3). SLA was significant (P ≤0.05) for 3 GSs at the level of ***p<0.001. Adjusted R$^2$ for SLA was 0.083, 0.352 and 0.486 at 20, 30 (DAS) and harvest respectively. Adjusted R$^2$ was 0.378 for SLA mean of 3 GSs.
**SLW (g cm$^{-2}$)**

Specific Leaf Weight was only significant crop and system-wise at 30 DAS and harvest. SLW was higher in S$_1$ compared to S$_2$. Interactions were insignificant (P ≤0.05). Adjusted R$^2$ for SLW was 0.105, 0.262 and 0.440 at 20, 30 (DAS) and harvest respectively (Fig. 4). Adjusted R$^2$ was 0.399 for SLW of the mean of 3 GSs. SLW was also significant for 3 GSs at the level of **p<0.001.** SLW increased over 3 GSs.

**NAR (g g$^{-1}$ t$^{-1}$)**

Net Assimilation Rate significantly higher in S$_1$ compared to S$_2$. NAR was significant at the level of **p<0.001 for (S$_{1/2}$ : NM$$_{1-5}$) and (S$_{1/2}$ : C$_{1-3}$) interactions. NAR steadily increased over 3 GSs for all 3 crops (Fig. 5). NAR of C$_3$ was significantly higher compared to C$_1$ and C$_2$. Adjusted R$^2$ for NAR was 0.266 and 0.419 at 30 (DAS) and harvest respectively. Adjusted R$^2$ was 0.306 for NAR of the mean of 3 GSs.

**Total DW (gm) of the plant**

Total dry weight (DW) of all 3 crops (C$_1$, C$_2$ and C$_3$) is significantly higher in S$_1$ compared to S$_2$ (Fig. 2). Treatments- and system-wise total DW increased significantly over the growth stages, however, interaction between system: treatment within same system (S$_{1/2}$ : NM$$_{1-5}$) remained insignificant (Fig. 6). Total DW of C$_3$ was significantly higher than C$_1$ and C$_2$. System-wise and treatment-wise total DW was significant at the level of **p<0.001 for all 3 GSs. Significant difference in total DW between S$_1$ and S$_2$ at the level of **p<0.001 at 20 and 30 DAS. Total DW increased significantly (P ≤0.05) over the growth stages. (S$_{1/2}$ : NM$$_{1-5}$) interaction was not significant. Adjusted R$^2$ for total DW was 0.467, 0.603 and 0.581 at 20, 30 (DAS) and harvest respectively. Adjusted R$^2$ was 0.525 for total DW of mean of 3 GSs.
Figure 5. Changes in mean of NAR (g g\(^{-1}\) t\(^{-1}\)) with time. (S\(_{1/2}\); NM\(_{1-5}\)) interactions at 20, 30 and 40 DAS (harvest), where 1\(^{st}\) row is for C\(_1\) (1\(^{st}\) two plots) and C\(_2\) (2\(^{nd}\) two plots); 2\(^{nd}\) row is for C\(_3\) (1\(^{st}\) two plots) and mean of 3 crops (2\(^{nd}\) two plots).

Figure 6. Changes in mean of total DW (gm) with time: (S\(_{1/2}\); NM\(_{1-5}\)) at 20, 30 and 40 DAS (harvest) where 1\(^{st}\) row is for C\(_1\) (1\(^{st}\) two plots) and C\(_2\) (2\(^{nd}\) two plots); 2\(^{nd}\) row is for C\(_3\) (1\(^{st}\) two plots) and mean of 3 crops (2\(^{nd}\) two plots).

DISCUSSION

The influence on the growth condition of protected systems was significant. LAR, LWR, SLA and total DW. NAR was significant overgrowth period with system interaction. Except for NAR and total DW, effect of organic treatments on other growth parameters was not significant (>\(P \leq 0.05\)). However, effect of organic treatments on total DW production was significant. A similar effect was reported by (Kumar & Arumugam 2010) under nitrogen treatment in sunflower. There was significant effect on total DW including on other growth parameters as a result of system:treatment interactions (Fig. 6, Table 3). Treatment with vermicompost (NM\(_2\)), followed by vermicompost+neem cake (NM\(_3\)) and compost (NM\(_5\)) resulted to higher total DW production irrespective of system influence. Crops response to organic treatments combined with rock phosphate (NM\(_4\)) was poor. This could be because of the fact that effectiveness of rock phosphates as fertilizer depends on soil characteristics, especially acidity, and calcium and phosphate status. If these above mentioned soil characteristics are not conducive, effect of rock phosphates on short-lived herbaceous plants such as lettuce, spinach etc. will be minimal (Bolland & Gilkes 1990, Le Mare 1991) because of slow solubilization process.
NAR and total DW production was significantly higher in polyhouse (S1) compared to shadenet (S2) which increased proportionately over the growth stages. The result on growth parameter study also shows that there was decrease in LWR and SLA which could have been affected by spacing between the plant rows (Papadopoulos & Pararajasingham 1997).

CONCLUSIONS
Growing vegetables in protected system like in different types of polyhouse and shadenet (color and shade factor) have been reported to optimize the agricultural productivity both in terms of quality and quantity. Comparative study on growth parameters between two different protected systems and interactions between them complements such findings. Higher growth parameter values in polyhouse and it’s variation during growth stages in green leafy vegetables grown not only attributed increased in DW of the vegetables but also indicated better production than in shadenet. Green leafy vegetables in polyhouse have performed significantly better than vegetables in shadenet. Total DW of all 3 leafy vegetables was less influenced by organic treatments compared to system: crop interaction. Care should be taken to collect preliminary report on the soil nutrient status, previous crops’ pest and disease history and cropping system as to appropriately study the multifactorial influence on DW production under protected cultivation so that extraneous influence is minimized.

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