

GROWTH ENVIRONMENT AND POT VOLUME AFFECT BIOMASS AND ESSENTIAL OIL PRODUCTION OF BASIL¹

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ABSTRACT – The objective of this work was to evaluate the effect of pot volume and growth environment on the productions of biomass and essential oil of basil plants (*Ocimum basilicum* L.). A completely randomized experimental design was used, with five replications, in a 6×2 factorial arrangement consisting of 6 growth environments (full sun; 50% black shade screen; 50% silver shade screen; 50% red shade screen; 35% green shade screen; 150 µm low density polyethylene film - LDPE) and two pot volumes (3.5 L and 5.0 L). The plants were cut and evaluated for variables related to growth, root system, and extraction of essential oil. The growth environments and pot volumes affected the production of biomass and essential oil of the basil plants evaluated. Plants grown under red and silver shade screens had 36.03% and 31.31% higher plant height than those grown at full sun, respectively. Basil plants grown in 5.0-liter pots under black shade screen produced higher essential oil contents. The biomass production of basil plants grown in 5.0-liter pots was affected by the red and green shade screens and LDPE film. The growth of basil plants in 5.0-liter pots under 50% black shade screen is recommended when the crop is intended for essential oil extraction; and their growth in 5.0-liter pots under red shade screen, green shade screen, or LDPE film is recommended when the crop is intended for fresh biomass production.

Keywords: Aromatic plant. Shading. Colored screens.

AMBIENTES DE CULTIVO E VOLUME DO VASO INFLUENCIAM A PRODUÇÃO DE BIOMASSA E ÓLEO ESSENCIAL DE MANJERICÃO

RESUMO - Objetivamos avaliar a influência de volumes de vasos sob diferentes ambientes de cultivo no crescimento vegetativo e a produção de óleo essencial de manjeriço (*Ocimum basilicum* L.). O delineamento experimental utilizado foi o inteiramente casualizado, disposto em esquema fatorial 6x2, com cinco repetições. O primeiro fator foi os ambientes de cultivo (pleno sol, tela de sombreamento preta 50%, tela de sombreamento prata 50%, tela de sombreamento vermelha 50%, tela de sombreamento verde 35% e filme polietileno de baixa densidade 150 µm - PEBD) e o segundo fator foi o volume de vaso (3,5 e 5,0 L). Foi realizado o corte das plantas e a avaliação das variáveis de crescimento, de sistema radicular e extração de óleo essencial. O ambiente de cultivo e o tamanho do vaso influenciam na produção de biomassa e de óleo essencial de manjeriço. As plantas cultivadas sob as telas vermelha e prata tiveram incrementos de 36,03% e 31,31%, respectivamente, na altura das plantas em relação ao cultivo em pleno sol. Plantas de manjeriço cultivadas em vasos de 5,0 L sob tela preta produziram maior teor de óleo essencial. O crescimento vegetativo de plantas de manjeriço cultivadas em vasos de 5,0L foi influenciado pelas telas vermelha, verde e filme PEBD. Considerando a produção de manjeriço para extração de óleo essencial recomenda-se o cultivo em vasos de 5,0 L sob tela preta, e se a exploração for destinada a produção de biomassa fresca, orienta-se pelo cultivo com o mesmo vaso sob telas vermelha, verde ou filme PEBD.

Palavras-chave: Planta aromática. Sombreamento. Telas coloridas.

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INTRODUCTION

Basil (*Ocimum basilicum* L.) is native to Asia (Middle East) and belongs to the Lamiaceae family; it is an annual or perennial plant depending on the location that it is grown (LUZ et al., 2014). Basil is commercially grown for use of its leaves fresh or dried as aromatizing and condiment products, and for essential oil extraction, which is important for perfumery, food, and beverage industries (BLANK et al., 2007). According to Ereno (2006), basil essential oil is composed of 40.2% to 48.5% linalool, which is used as a perfume fixative.

The production of medicinal plants has a global importance due to the demand of chemical, pharmaceutical, food, and cosmetic industries (SOUZA et al., 2013). According to Blank et al. (2007), basil has great agronomical potential for linalool extraction because of its oil content and short cycle.

The world basil production is approximately 830 Mg year⁻¹, and the extraction of its essential oil generates an income of US\$ 6.5 million. Although data for this crop in Brazil is scarce, it is the second most marketed aromatic plant in the Warehouse and General Storage Company of São Paulo (CEAGESP), with volumes of 5,000 to 7,000 bunches per month; and the varieties with purple leaves represent 15% to 20% of the total basil produced in Brazil (GENUNCIO et al., 2018).

The environment and crop management affect the growth, production of biomass, and chemical composition of essential oils of medicinal plants (CHAGAS et al., 2013).

Solar radiation is the main factor that limits yield of crop species grown at field conditions or under protected environments, and morphophysiological responses of crop species may vary when the light intensity is altered according to their capacity of acclimation and dependence on light quantity or quality (LIMA et al., 2008).

Colored shade screen is a new agrotechnological concept to combine physical protection and solar radiation filtration, promoting specific physiological responses that are regulated by light (BRANT et al., 2009). Thus, changes in spectral characteristics of solar radiation may modify structural and physiological characteristics of plants (SOUZA et al., 2014).

Another factor that affects medicinal plant production is the pot volume for plant growth, mainly under protected environments or when adopting domestic crops for growing in limited physical spaces (apartments, balconies, vertical crops); thus, the determination of the pot volume for each species is important.

Small pots can limit plant development because of their low volume, physically limiting root growth (ALMEIDA et al., 2014). The pot volume affects plant yield, phenology, and architecture, since

large pots (4.0 and 6.0 L) can increase plant branching, and small pots (1.0 and 3.0 L) can anticipate the flowering stage of some species (CAMPOS; MEDONÇA, 2013).

Considering the current importance of basil crop and that the growth management used can affect the productions of biomass and essential oil of this plant species, the objective of the present work was to evaluate the effect of the pot volume and growth environment on the production of biomass and essential oil of basil plants.

MATERIAL AND METHODS

The experiment was conducted from November 2018 to February 2019 at the experimental area of the Federal University of Western Bahia, Barra Multidiscipline Center, Barra, BA, Brazil (11°5'23"S, 43°8'30"W, and average altitude of 398 m). The average, minimum, and maximum temperatures during the experiment period were 26.5, 20.8, and 32.1 °C, respectively, and the accumulated rainfall was 352 mm. The experiment was conducted in a completely randomized design with five replications, using a 6×2 factorial arrangement consisting of 6 growth environments (full sun; 50% black shade screen; 50% silver shade screen; 50% red shade screen; 35% green shade screen; and 150 µm low density polyethylene film - LDPE) and two pot volumes (3.5 L and 5.0 L).

The growth environments were 2.0 m wide and 6.0 m long (12 m²) and covered by shade screens. The pots were randomly placed in four rows, with a spacing of 0.60 × 0.40 m (41,666 plants ha⁻¹) inside each environment.

Basil seeds of the cultivar Folha Fina (Topseed®) were sowed in 72-cell expanded polystyrene trays filled with substrate based on organic compost, using four seeds per cell, for production of seedlings, which were thinned at 15 days after emergence of the plants. The seedlings were transplanted to the pots at 30 days after emergence, when they had 6 true leaves. The physical and chemical characteristics of the soil used for growing the basil plants are described in Table 1.

The soil in the pots was fertilized at planting using 1 L of a nutritive solution per liter of soil. This solution used was based on Hoagland and Arnon (1950) and consisted of 12.0 mmol dm⁻³ KNO₃ (1.21 g dm⁻³), 2.0 mmol dm⁻³ NH₄H₂PO₄ (0.23 g dm⁻³), 4.0 mmol dm⁻³ MgSO₄·7H₂O (0.48 g dm⁻³), 8.0 mmol dm⁻³ Ca(NO₃)₂ (1.31 g dm⁻³), 0.6 µmol dm⁻³ CuSO₄·5H₂O (0.1 mg dm⁻³), 2.6 µmol dm⁻³ ZnSO₄·7H₂O (0.75 mg dm⁻³), 92 µmol dm⁻³ H₃BO₃ (5.7 mg dm⁻³), 25.2 µmol dm⁻³ MnCl₂·4H₂O (5.0 mg dm⁻³), 0.2 µmol dm⁻³ (NH₄)₆Mo₇O₂₄·4H₂O (0.25 mg dm⁻³), and 90 µmol dm⁻³ FeSO₄·7H₂O-EDTA bisodium salt (25.02 mg dm⁻³). Topdressing

consisted of 120 kg ha⁻¹ of N (2.88 g plant⁻¹) and 100 kg ha⁻¹ of K₂O (2.40 g plant⁻¹), divided into three applications (30%, 30%, and 40% at 20, 35, and 50 days after transplanting to the pots, respectively).

Weeds were controlled at their emergence by removing them from the pots; pests and diseases were monitored daily and no pest or disease were found over the experiment period.

Table 1. Physical and chemical characteristics of the soil used for the basil (*Ocimum basilicum* L.) crop.

pH (H ₂ O)	P		Ca ²⁺	Mg ²⁺	H+Al	CECe	CEC	SB	BS	AS	OM	Sand	Silt	Clay
	mg dm ⁻³													
5.5	0.9	46	0.9	0.20	1.16	1.24	4.19	1.24	57	0	1.24	81	10	9

H + Al = potential or total acidity; CEC = cation exchange capacity at pH 7.0; CECe = effective CEC; SB = sum of bases; BS: base saturation; AS: aluminum saturation; OM = organic matter.

The plants were harvested at 60 days after transplanting, when the plants were at the beginning of the flowering stage. The plants were cut and evaluated for variables related to growth, root system, and extraction of essential oil. Plant height, stem and canopy diameters, plant length, and root system volume were evaluated. The root system volume was obtained by the method of displacement of water in a graduated cylinder. The root system density was obtained through the relation between root fresh weight and root volume. The leaf area was obtained using a measurer device for leaf area (Li-3100; LiCor, Lincoln, USA).

The plants were separated in leaves, stems, and roots, weighed using an analytical balance, and quantified for leaf fresh weight, stem fresh weight, root fresh weight, total fresh weight, shoot fresh weight (leaf and stem fresh weights), and root to shoot ratio. The yield corresponded to the total weight of leaves harvested from plants of each plot.

The hydrodistillation method was used for leaf essential oil extraction. Leaf samples of 100 g were used for each experimental unit, which were placed in a extractor device (Vidrolabor Clevenger; Labor Quimi, Poá, Brazil) where the leaves were in contact with 1.0 L of water at 105 °C. The essential oil was volatized with water vapors and condensed in a closed system, where the oil layer was separated from the aqueous stage. Each extraction of basil oil had 90 minutes, as previously established for oil extraction of this species. Then, the volumes were read and the essential oil yields (L ha⁻¹) were calculated.

The data collected were subjected to analysis of variance and the means were compared using the Tukey's test at 5% probability, in the Sisvar 5.6 program (FERREIRA, 2011).

RESULTS AND DISCUSSION

The interaction between the factors (growth environment and pot volume) was significant for root fresh weight, leaf fresh weight, total fresh weight, leaf area, oil content, and oil yield (Table 3). The effect of the growth environment factor was significant for plant height, root length, root system

density, and root to shoot ratio (Table 2). The effect of the pot volume factor was significant for plant height, stem diameter, canopy diameter, root length, root system density, and root to shoot ratio (Table 2).

The plants grown under the 50% red and 50% silver shade screens had 36.03% and 31.31% higher plant heights, respectively, than those grown under full sun conditions (Table 2). This result was probably due to a higher transmission of far-red wavelengths by the red screen. According to Taiz and Zeiger (2015), the higher the far-red radiation, the higher the stem elongation rate in heliophyte species, and this elongation characterizes a response of the plant to avoid shading to capture more light energy, indicating the involvement of the phytochrome with shade perception.

The growth environment factor had no effect on stem diameter and canopy diameter (Table 2). However, despite the lack of significant differences, all covers tend to increase these variables, except the 50% silver shade screen for stem diameter, and the 50% black shade screen for canopy diameter.

Souza et al. (2014) found lower stem diameter for *Rosmarinus officinalis* plants grown under red or blue screen when compared to those grown under full sun conditions. Martins et al. (2008) found that *Ocimum gratissimum* L. plants grown at full sun had higher stem base diameter than those grown under colored screens. Reductions in light intensity result in plants with longer stems and lower canopy expansion because these characteristics present great plasticity between plant species and are affected by the crop conditions (TAIZ; ZEIGER, 2015). Thus, the environments that favored solar radiation availability for the basil plants evaluated in the present work contributed to the increases in canopy and stem diameters, because the rapid leaf expansion favored the maximum capture of light and conversion into biomass by the plants; a large stem diameter is a desirable characteristic because it generates a better support for the plant shoot system.

The basil plants grown under the 50% black shade screen or 50% silver shade screen had higher root system length than those grown under full sun conditions; and the plants grown under 50% silver shade screen had higher root system density than

those grown at full sun (Table 2). The plants grown under full sun or LDPE film environments had higher root to shoot ratio than those grown under the

other environments, indicating a better root system growth under these crop conditions (Table 2).

Table 2. Plant height, stem diameter, canopy diameter, root to shoot ratio (R/S), root length, and root system density of basil plants (*Ocimum basilicum* L.) grown under different environments and pot volumes.

Growth environment	Plant height (cm)**	Stem diameter (mm) ^{ns}	Canopy diameter (cm) ^{ns}	Root length (cm)**	Root system density (g cm ⁻³)**	R/S**
Full sun	39.88 c	7.75 a	30.44 a	19.51 c	0.21 b	1.44 a
50% black shade screen	44.50 b	7.53 a	24.50 a	29.50 a	0.17 b	0.63 c
50% silver shade screen	52.37 a	6.54 a	28.75 a	29.94 a	0.30 a	0.77 b
50% red shade screen	54.25 a	7.10 a	30.00 a	27.63 ab	0.23 ab	0.91 b
35% green shade screen	40.45 c	7.41 a	28.84 a	26.94 ab	0.25 ab	0.91 b
150 µm LDPE film	46.17 b	7.08 a	31.54 a	22.98 bc	0.24 ab	1.13 a
MSD	4.69	1.84	7.76	5.78	0.08	0.48
Pot volume						
3.5 L	44.35 b	6.30 b	24.41 b	23.03 b	0.21 b	1.07 a
5.0 L	48.19 a	8.18 a	33.61 a	29.13 a	0.26 a	0.87 b
MDS	1.82	0.71	3.02	2.24	0.03	0.19
CV (%)	6.75	16.91	17.78	14.73	27.60	23.60

Means followed by the same letter in the columns are not different by the Tukey's test at 5% probability. **, *, and ns = significant at 1% probability, 5% probability, and not significant by the F test. LDPE = low density polyethylene. MSD = minimum significant difference. CV = coefficient of variation.

Martins et al. (2008) evaluated the response of basil plants (*O. gratissimum* L.) to growth environments and found that root biomass accumulation varies depending on the shade screen used. Brant et al. (2009) evaluated *Melissa officinalis*, and Souza et al. (2011) evaluated *Mikania glomerata* Spreng. plants; they found more accumulation of dry biomass in plants that grew under colored screens than in those that grew under full sun conditions. According to Larcher (2004), shaded plants distribute less dry biomass to roots, indicating an adaptative response that provides higher carbon gains, such as increases in leaf area, or a strategy to search for light. The results found by Martins et al. (2008) confirm those found in the present study; they found higher root to shoot ratio in crops grown under full sun, indicating a higher biomass allocation to the root system when compared to plants grown under shading.

The use of a 5.0-liter pot to grow the basil plants had positive effect on plant height (+9.81%), stem diameter (+31.88%), canopy diameter (+38.55%), root length (+27.48%), and root system density (+23.86%); and the use of a 3.5-liter pot had positive effect only on root to shoot ratio (+22.98%) (Table 2).

The results showed that the use of the 5.0-liter pot favored the growth of basil shoot and root systems due to the greater availability of space with substrate volume. According to Poorter et al. (2012), the low plant growth in 1.0-liter pots is caused mainly by decreases in photosynthesis per unit of leaf area, which causes a stress to the plant, limiting its root system and, consequently, its shoot growth. Campos and Mendonça (2013) evaluated the effect

of pot volume on sweet basil development and found similar results; the plants grown in 4.0- and 6.0-liter pots had mean growth rates of 1.0 and 1.2 cm day⁻¹, whereas plants grown in 1.0- and 3.0-liter pots had means of 0.9 and 0.6 cm day⁻¹, respectively; they concluded that the stress caused by the smaller pots affected the plant architecture.

The interaction between the factors (growth environment and pot volume) was significant for shoot fresh weight, root fresh weight, and total fresh weight. Plants grown in 5.0-liter pots under full sun conditions or 50% red shade screen presented higher root fresh weight. The plants grown in 5.0-liter pots under any of the growth environments, except that with 50% silver shade screen, presented increases in leaf fresh weight. The total fresh weight of plants grown in 5.0-liter pots under all the growth environments increased, except that of plants grown under 50% black or 50% silver shade screens (Table 3).

The explanation for the higher growth of basil plants under higher substrate volumes is based on the findings of Brum et al. (2007), who found that increases in diameter of *Crisantemo multiflora* plants were proportional to increases in volume of pots; they explained these results by the restriction of the root system development and reductions in biosynthesis and translocation of cytokinin, gibberellin, and specific amino acid from roots to shoots when using low-volume pots, which decrease leaf expansion and branch growth (TAIZ; ZEIGER, 2015). Moreover, Poorter et al. (2012) reported that the proper pot volume for the best plant performance will depend on the size of the plant that will grow on it and, based on several recommendations, they

indicated the use of pot volumes that do not allow the plant to produce more than 1 g L⁻¹ of biomass.

Plants grown in 5.0-liter pots and under environments with red shade screen, green shade screen, 150 µm LDPE film, or at full sun had higher leaf area (Table 3). Increases in leaf area of plants grown under shading may be a plant strategy for capturing of light and maximization of the use of light rays. Larcher (2004) explains that heliophyte plants efficiently use high radiation intensities because of the high capacity of their electron

transport system; therefore, they have higher photosynthetic gains and biomass accumulations. This explains why the use of red shade screen resulted in basil plants with higher efficiency for biomass accumulation than those that grew under the other treatments. Martins et al. (2008) evaluated *O. gratissimum* plants and found that the use of colored screens provided higher biomass accumulations than the black screen, showing that this pattern repeats in different studies.

Table 3. Root fresh weight, leaf fresh weight, total fresh weight, leaf area, oil content, and oil yield of basil plants (*Ocimum basilicum* L.) grown under different environments and pot volumes.

Growth environment	Root fresh weight (g)**		Leaf fresh weight (g)*	
	Pot volume			
	3.5L	5.0L	3.5L	5.0L
Full sun	30.67ABb	79.04Aa	12.89Bb	35.64Aa
50% black screen	14.69Ba	19.61Ca	14.81ABb	30.20Aa
50% silver screen	21.73ABb	41.64Ba	22.77ABa	25.89Ba
50% red screen	31.89ABb	68.74Aa	22.39ABb	46.42Aa
35% green screen	35.83ABa	48.19Ba	30.24Ab	42.39Aa
150 µm LDPE film	45.63Aa	47.67Ba	22.78ABb	43.43Aa
MSD (pot)	18.56		11.36	
MSD (environment)	27.54		16.86	
CV (%)	22.00		17.19	
	Total fresh weight (g)**		Leaf area (m ²)**	
	Pot volume			
	3.5L	5.0L	3.5 L	5.0 L
Full sun	53.83Ab	141.58Aa	0.032 Bb	0.125 ABa
50% black screen	37.85Bb	67.35Ca	0.052 ABa	0.100 Ba
50% silver screen	62.30Ab	95.79Ba	0.125 Aa	0.092 Ba
50% red screen	69.43Ab	153.54Aa	0.082 ABb	0.168 ABa
35% green screen	71.61Ab	124.48Aa	0.108 ABb	0.190 Aa
150 µm LDPE film	84.73Ab	120.06Aa	0.049 ABb	0.169 ABa
MSD (pot)	23.65		0.057	
MSD (environment)	35.09		0.084	
CV (%)	18.28		18.28	
	Oil content (mL 100 g ⁻¹)**		Oil yield (L ha ⁻¹)**	
	Pot volume			
	3.5L	5.0L	3.5 L	5.0 L
Full sun	0.51 Aa	0.23 Bb	21.22 ABa	9.67 Bb
50% black screen	0.25 Bb	0.52 Aa	10.45 CDb	21.74 Aa
50% silver screen	0.29 Ba	0.33 ABa	12.34 BCDA	13.70 ABa
50% red screen	0.21 Ba	0.24 Ba	8.83 Da	10.07 Ba
35% green screen	0.44 ABa	0.12 Bb	18.49 ABCa	5.00 Bb
150 µm LDPE film	0.61 Aa	0.26 Bb	25.45 Aa	10.83 Bb
MSD (pot)	0.14		6.24	
MSD (environment)	0.22		9.26	
CV (%)	21.10		21.13	

Means followed by the same uppercase letter in the columns or lowercase letter in the rows are not different by the Tukey's test at 5% probability. ** and * = significant at 1% and 5% probability by the F test. LDPE = low density polyethylene. MSD = minimum significant difference. CV = coefficient of variation.

Corrêa et al. (2012) evaluated the productive performance of *Origanum vulgare* plants under colored screens environments and under full sun conditions and found that the plants were sensitive to light quality, since the plants under blue light produced lower roots and lower total dry biomass than those under red and black screens. Probably, basil plants are also sensitive to light quality, because different fresh weight accumulations were found for different parts of the plants when they grew under different wavelengths.

Oliveira et al. (2009) found higher leaf area in *Artemisia vulgaris* L. plants grown under shade screens, and they attributed this result to a plant strategy to maximize light capture. According to Larcher (2004), higher total leaf area, specific leaf area, and leaf area ratio indicate higher photosynthetic surface, lower leaf thickness, and higher proportion of photosynthetically active tissues. Moreover, Jones and Mcleod (1990) reported that the use of this strategy by the plant to increase the photosynthetic surface ensures a more efficient use of low light intensities under shading and, consequently, compensates low photosynthetic rates per unit of leaf area, which is a characteristic of shaded leaves.

The plants grown in 5.0-liter pots under 50% black or 50% silver shade screen presented higher essential oil contents and yield than those grown in the other environments. When the basil plants were grown in 3.5-liter pots under LDPE film or at full sun, they presented higher essential oil contents and yield (Table 3).

The basil plants had no positive response to the use of some shade screens, depending on the pot volume, for essential oil production. The higher essential oil contents found in plants grown in 3.5-liter pots at full sun or under LDPE film can be related to a strategy of adaptation of the plants to these environments, which affect their root growth, air temperature, wind, irradiance incidence, and, consequently, water demand and other environmental factors. According to Brant et al. (2009), changes in the wavelength on plants promoted by red, black, and blue screens cause a lack of some light wavelengths that are not absorbed by plant pigments because of the filtering of the screens, which may hinder some important routes of terpene synthesis and the production of essential oil. These plants present higher essential oil production under stress conditions due to changes in the biosynthesis routes (LUZ et al., 2014), which explains the increase in basil essential oil contents when the plants were grown under full sun or LDPE film environments, with restrictions for root growth (in 3.5-liter pots).

The basil plants grown in 5.0-liter pots under 50% black shade screen had higher essential oil production, but lower leaf fresh weight and leaf area, denoting a high oil accumulation in shading conditions. According to Taiz and Zeiger (2015),

plants grown under shading develop thinner leaf blades than plants grown under full sun conditions, and this can be a mechanism of protection of plants against high solar radiations. Similarly, Chagas et al. (2013) found that *Mentha arvensis* L. plants grown under 50% shade screens had changes in leaf and growth characteristics, denoting their phenotypical plasticity when they were grown in environments under different lights; the greater effect of light intensity than light quality on these plants; and that synthesis and accumulation of essential oil can occur under shaded, highly lit environments; however the plants in the treatments under shading had greater leaves with lower oil concentration. Gomes et al. (2009) found that *Lippia citriodora* plants grown under full sun conditions had greater biomass production, and those under 70% shading had higher number of trichomes.

Martins et al. (2008) reported that the blue light contributed to a higher essential oil content in *O. gratissimum* plants grown under blue screen when compared to those grown under red screen or at full sun. Thus, basil plants may respond differently to environmental stimuli for production of biomass and essential oil.

CONCLUSION

Basil plants grown in 5.0-liter pots under a 50% black shade screen had higher production of essential oil content.

The biomass production of basil plants grown in 5.0-liter pots was affected by the growth environments with 50% red shade screen, 50% green shade screens, and LDPE film.

The growth of basil plants in 5.0-liter pots under 50% black shade screen is recommended when the crop is intended for essential oil extraction, and their growth in 5.0-liter pots under red shade screen, green shade screens, or LDPE film is recommended when the crop is intended for fresh biomass production.

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