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Supplemental irrigation levels in bell pepper under shade mesh and in open-field: absolute growth rate, dry mass, leaf area and chlorophyll

Richard Alberto Rodríguez Padrón[1]*, Jerson Vanderlei Carús Guedes[2], Alexandre Swarowsky[3], Cicero Urbanetto Nogueira[4], Roxanna Rosales Cerquera[4] and Helena Maria Camilo de Moraes Nogueira[4]

[1]Center of Rural Science, Federal University of Santa Maria, Campus Camobi, Santa Maria, RS, Brazil.
[2]Department of Phytosanitary Defense, Federal University of Santa Maria, Santa Maria, RS, Brazil.
[3]Department of Environmental Engineering, Franciscan University, Santa Maria, RS, Brazil.
[4]Polytechnic School of the Federal University of Santa Maria, Santa Maria, RS, Brazil.

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This study aims to evaluate the effect of supplemental irrigation levels on vegetative parameters of bell pepper grown in open field and under shade mesh. The experimental design was a randomized complete block with four replications and ten treatments in factorial scheme (four irrigation levels combined with shade). Irrigation treatments consisted in 0.25, 0.50, 0.75 and 1.0 rate of crop evapotranspiration and the control (no-irrigation).Shading treatment was of 50% reduction of the photosynthetically active radiation compared to open field conditions. Vegetative parameters were influenced by irrigation. The growth rate of plants present no significant difference. The growth rate of stem diameter present difference, being treatments 0.50 and 0.75 the highest.Significant interaction was present in the rate of chlorophyll, dry matter, leaf area index and number of leaves per plant. Plots under shade mesh showed the highest growth rate in plant height and stem diameter and leaf area index, number of leaves per plant, dry matter and lower chlorophyll index. As irrigation strategy, considering the water use efficient and vegetative characteristics of bell pepper, the most favorable irrigation levels were 0.5 and 0.75 of ETc, under shade and in open field, respectively, without affecting the vegetative parameters and yield.

Key words: *Capsicum annuum*, water stress, water use efficiency, drip irrigation.

INTRODUCTION

Bell pepper (*Capsicum annuum* L.) is a member of the Solanaceous family, native to Mexico, Central America and northern South America (Echer et al., 2002; Filgueira, 2003; Souza et al., 2011). It is an important crop in many parts of the world, given their economic importance, ranking second in world production. It is

Author for correspondence. E-mail: rarpadron@gmail.com/richardpadron@mail.ufsm.br, Tel.: (+55) 5532208158.

Table 1. Monthly climatic data of the experimental area, relative humidity, insolation and evaporation cumulative during the seasons 2013-2014 and 2014-2015.

Month	Relative humidity mean (%)		Insolation (hour)		Evaporation (mm)	
	2013-2014	2014-2015	2013-2014	2014-2015	2013-2014	2014-2015
Nov	71.60	71.0	229.2	173.1	144.5	131.8
Dec	69.45	76.1	286.2	211.0	175.2	142.2
Jan	73.15	78.3	219.2	212.4	158.3	142.0
Feb	73.79	79.7	211.2	218.0	137.9	123.9
Mar	76.87	77.6	212.6	208.4	112.8	114.1
Sum	-	-	1376.8	1189.6	850.8	804.7

considered one of the ten species of greatest economic importance in the Brazilian vegetable market, and the area annually cultivated is around 13 mil hectares, with production close to 290 mil tons of fruit, generally grown in open field (Marouelli and Silva, 2012).Water use by plants and all the physiological processes are directly related to their status in the soil-water-plant system.The interrelationships between these factors are fundamental for the planning and the operation of irrigation systems to maximizeyield and product quality (Trani and Carrijo, 2004). There are many motivations to study the physiology of plants under stress, among which: knowledge of stressors on plants can be crucial for the development of mechanistic models predictive in nature, for example, the study of the possible effects of climate change. The analysis of the interaction of the plants with the environmental factors are fundamental to comprise the distribution of the species in the different ecosystems and the performance of the crop is strongly limited by the impact of the environmental stress (Nilsen and Orcutt, 1996).

Increased temperatures occurring in late spring and early summer reduce bell pepper yields and increase incidence of physiological disorders in fruit, such as blossom-end rot and sunscald (Olle and Bender, 2009). High temperatures induce flower abortion and fruit in bell pepper (Deli and Tiessen, 1969). Bell pepper is a very demanding plant in luminosity, especially in the early stages of reproduction (Prieto et al., 2003). The increment in crop production is able to be possible only knowing the pushing effects of irrigation and radiation on plant growth and yield (Kara and Yildirim, 2015). The amount of solar radiation intercepted by plants is a major determinant for the total dry matter produced by a crop (Biscoe and Gallagher, 1978). The most effective development forces on plants are “Carbon”, “Water”, “Radiation” and energy supply of plants’ comes from radiation also (Steduto, 2003). Plant development depends on the amount of radiation, duration of light in a day, relative humidity, wind speed and temperature (Boztok, 2003). Also, plant water, nutrient uptake and transpiration rate are closely related with solar radiation (Adams, 1992).

The analysis of plant growth allows evaluating the behavior of crop genotype in relation to different crop systems, influenced by management, climate and crop physiology (Oliveira et al., 2015). The indexes wrapped in the analysis of growth, as foliar area index, growth rate, and liquid assimilation indicate the capacity of plants assimilatory system in synthesize and destine the organic matter in the diverse organs (Monte et al., 2009; Silva et al., 2010).That depend on photosynthesis, breathing and assimilate translocation of carbon fixation sites for local use and storage, where there are growth and differentiation of organs (Lopes et al., 2011).

Studying water requirements of plants has become increasingly important for agriculture, mainly for areas with deficit of irrigation water. This study aims to evaluate the effect of supplemental irrigation levels on vegetative parameters of bell pepper grown in open field and under shade mesh.

MATERIALS AND METHODS

The field study was conducted in the experimental area of the Polytechnic School of the Federal University of Santa Maria, located at an altitude of 110 min the geographic coordinates 29°41'25"S, 53°48'42"W, during the Spring Summer seasons of 2013-2014 and 2014-2015. The soil is classified as typical dystrophic yellow argissolo, with a loam texture (Streck et al., 2008). The climate of the region, according to the Köppen classification is subtropical humid (Cfa). According to the National Institute of Meteorology (INMET), mean annual evaporation, temperature and rainfall range from 800 to 1200 mm, 18 to 20°C and 1450 to 1650 mm, respectively. In Table 1 we can see the summary of the mean monthly climate data during the period of the experiment. The monthly cumulative insolation and evaporation in 2013-2014 season were higher than those in 2014-2015, except mean relative humidity. Solar radiation, evaporation, rainfall and daily temperature during the experimental period are shown in Figure 1. The monthly mean solar radiations in 2013-2014 were higher than in 2014-2015. The daily mean temperatures in 2013-2014 were higher than those in 2014-2015 except December and March. The monthly maximum temperatures were higher in December, January and February. The rainfall cumulative was higher in 2013-2014 (892.8 mm) and 2014-2015 (834 mm); the maximum monthly rainfalls were in November and December, respectively. The monthly radiation in 2013-2014 was higher in December and January.

The experimental design was a randomized complete block with

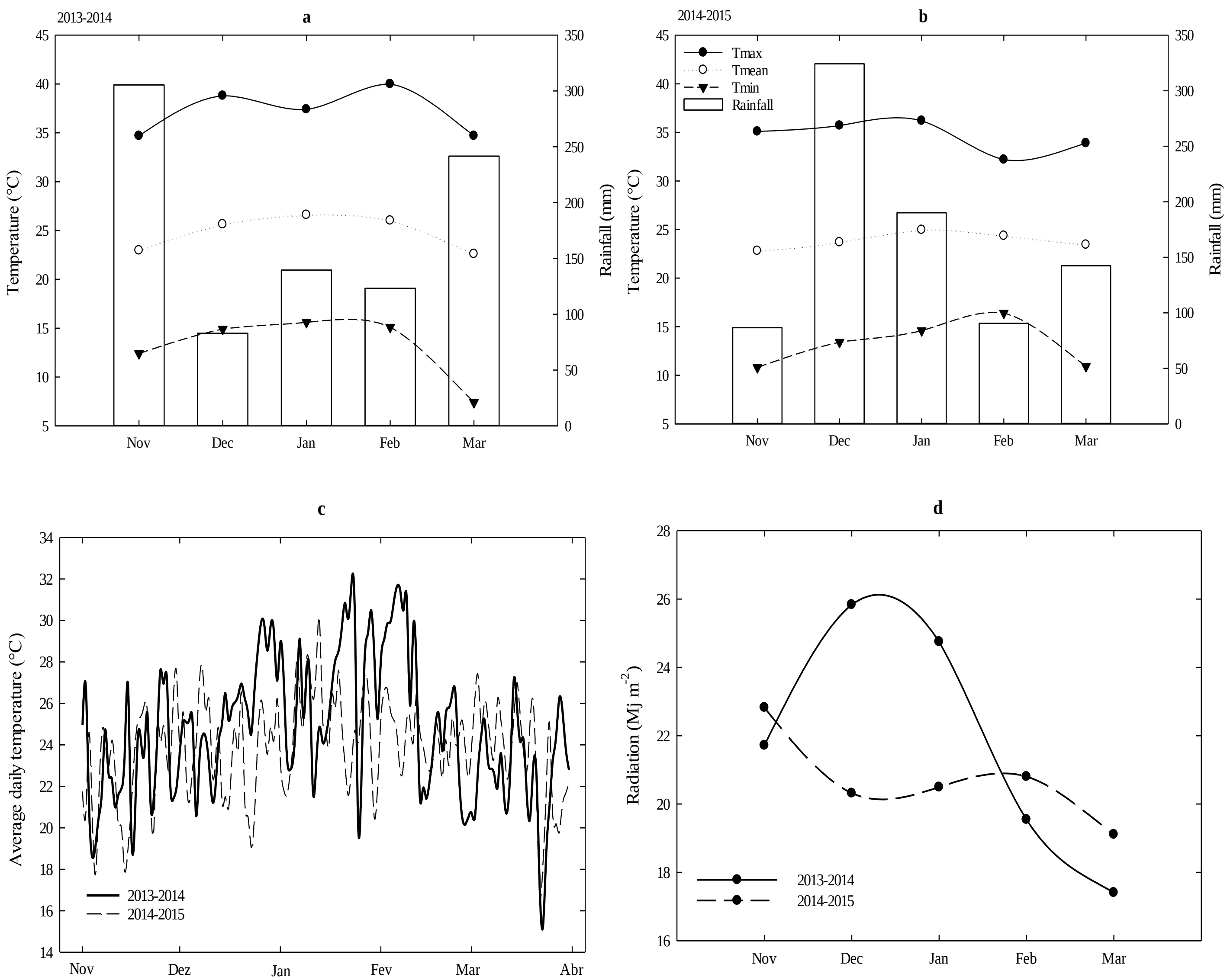

Figure 1. (a, b) Climograph of the experimental area, (c) average daily temperature and (d) radiation during the seasons 2013-2014 and 2014-2015.

four replications and ten treatments in factorial arrangement (four irrigation levels combined with shade mesh). Irrigation treatments were: 25% ($I_{0.25}$), 50% ($I_{0.50}$), 75% ($I_{0.75}$), and 100% ($I_{1.0}$) rate of crop evapotranspiration (ETc) and the control [no irrigation (I_0)].Shading treatments were 50% reduction of the photosynthetically active radiation (according to the manufacturer) and open field conditions (control, 0% shading). There were 40 experimental plots, each of 5.0 m long and 2 m wide (10 m^2), for a total area of 400 m^2, not including border plants. Moisture overlap between rows was avoided by border plants (1 m). Arcade was the variety of bell pepper used due to its commercial importance in the region. Plants were transplanted in the field with two months old at separation of 1.0 m between rows and 0.4 m between plants (density of 2.5 plantsm^{-2}), in Nov. 16, 2013 and Nov. 23, 2014. Shade mesh (polyethylene black shade mesh) was supported with metallic cable, in arectangular structure with the highest point at 2 m. The shade mesh was set two weeks before transplanting. The level of shading was verified by using digital radiometer (Model: MS-100). Leaf temperature was measured in each plot with an infrared thermometer gun (Model: AR 320)

It was used a localized irrigation system (drip) placed as lateral by row, spacing among emitters of 0.2 m and flow of 0.8 Lh^{-1}. It was installed in each experimental plot a ball valve for regulating the irrigation time and pressure regulating valve for uniformity. The irrigation strategy is described as follow: during the first 20 days after transplantation it was applied 100% of ETc to all treatments, to ensure in plants establishment. The levels of supplementary irrigation were applied from 20 to 119 days after transplanting and the frequency of daily watering was established. Due to the characteristics of soils and climatic conditions after the effective precipitation exceeds evaporation, irrigation was applied two days after with the frequency set.

The crop reference evapotranspiration (ETo) and crop evapotranspiration (ETc) were calculated using the following Equations (1) to (2).Use of reference evapotranspiration leads to increasing uncertainty comparing actual evapotranspiration. There

Table 2. Average soil parameters of the experimental area.

Soil layers (m)	Bulk density (g cm^{-3})	Field capacity (m^3 m^{-3})	Wilting point (m^3 m^{-3})	Water content (m^3 m^{-3})	Infiltration (mm h^{-1})	Texture
0-0.2	1.42	0.31	0.14	0.18		Loam
0.2-0.4	1.38	0.34	0.17	0.17	15.0	Clay-loam
0.4-0.6	1.36	0.37	0.23	0.13		Clay

are other models that can estimate evapotranspiration reference than have had successful results. Also, they are useful for selecting the best model when researchers must apply temperature-based models on the basis of available data (Valipour and Eslamian, 2014; Valipour, 2014a, b, c; Valipour, 2015a, b). Weather data was collected from an automatic weather station located 1 km from the experimental area. The crop reference evaporation (ETo) was calculated based on the method of FAO Penman-Monteith (Allen et al., 2006), (Equation 1) as follows:

$$\mathrm{ETo} = \frac{0.408\,\Delta\,(Rn - G) + \gamma\,\frac{900}{\mathrm{T}+273}\,U_2.(e_s - e_a)}{\Delta + \gamma\,(1 + 0.34\,U_2)} \quad (1)$$

Where ETo is reference evapotranspiration (mm day^{-1}), Rn, G and T are net radiation value at crop surface (MJ $m^{-2}day^{-1}$), soil heat flux density (MJ $m^{-2}day^{-1}$) and mean daily air temperature at 2 m height (°C), respectively. Also, u_2, e_se_a, (e_s- e_a), Δ and γ represent wind speed at 2 m height (ms^{-1}), saturation vapor pressure (kPa), actual vapor pressure (kPa), saturation vapor pressure deficit (kPa), slope of the saturation vapor pressure curve (kPa/°C) and psychrometric constant (kPa/°C), respectively.

The crop evapotranspiration (ETc) was calculated with the method of dual crop coefficient for each crop phonological stage (Allen et al., 2006), (Equation 2) as follows:

$$\mathrm{ETc} = (\mathrm{K_{cb}} + \mathrm{K_e}) \times \mathrm{ETo} \quad (2)$$

Where ETc crop evapotranspiration (mm), ETo reference crop evapotranspiration (mm) and splitting Kc into two separate coefficients, the basal crop coefficient Kcb and soil water evaporation coefficient Ke.

Before the plants were transplanted randomly sampling points were selected in the experimental area, to determine basic soil parameters, including soil texture, bulk density, field capacity, and permanent wilting point (Table 2). Also, it was performed an infiltration test to design the irrigation system.

The soil water content over the season was measured before and after irrigation every two days (four readings per experimental plot), with a portable time domain reflectometry (TDR-100). The two metallic sensor 0.2 m rods of the TDR were inserted vertically within the row between plants. Also monitoring was performed with neutron probe (CPN Model 503, DR), with calibration previous to execution of the experiment (Padrón et al., 2015a). PVC tubes (50 mm) were installed between row (1 m distance) and plant of each experimental plot at a depth of 0.7 m. Readings were performed once a week at 0.125, 0.30 and 0.50 m of soil depth.

Plant height and stem diameter was measured in ten plants per plot, sampled in a fifteen day interval. The absolute growth rate determination was done according to the formula described by Radford (1967). Equation (3), as follows:

$$\mathrm{AGR} = \frac{\mathrm{W_2} - \mathrm{W_1}}{\mathrm{T_2} - \mathrm{T_1}} \quad (3)$$

Where: AGR=absolute growth rate, W=mean plant height and mean stem diameter, T = time.

Leaf area and leaf number per plant was measured in three plants randomly selected per experimental plot, which were sampled in a fifteen-day interval. Eight samples during the experiment were determined with the application of model (LI-COR, Inc., USA). The leaf chlorophyll index was determined once a week over the season in five leaves per plant, in each plot, using a chlorophyll meter (ClorofiLOG, CFL1030, FALKER). Dry weight of the plant was performed at the end of the culture cycle in five plants per plot, randomly selected.The samples were determined separately (root and vegetative top). The plant samples were dried at 65°C until constant weight was obtained.

The main tasks of agricultural management were: weeding, weed control, insecticide and fungicide application and fertigation was according to the nutritional needs of the crop and chemical analysis of the soil. Fertigation was performed with irrigation (daily) at an irrigation rate of 40 t ha^{-1}. All plants received 368 kg ha^{-1} of a complete fertilizer (13N-14P_2O_5-13K_2O), 290 kg h^{-1} of ammonium nitrate (36% N) and 396 kg ha^{-1} of potassium nitrate (35% K_2O). The statistical analysis was performed using the SPSS software package (SPSS V17.0). Significant differences between means for different treatments were compared using Tukey test at P<0.05. Data from all years were pooled if no treatment interactions were found.

RESULTS AND DISCUSSION

The absolute growth rate of the plant height and diameter of stem for each treatment are shown in (Table 3). There was no significant interaction effect between treatments in open field and under shade mesh. On the other hand, the treatments under shade mesh showed higher growth rate. The stem diameter in open field and under shade mesh (P<0.05), the treatments $I_{0.75}$ and $I_{1.0}$ showed the highest growth rate in open field and under shade mesh $I_{0.25}$ and $I_{0.50}$. The treatments with lower growth rate were without irrigation.

The absolute growth rate of plant height and stem diameter during the crop cycle, for each treatment is shown in Figure 2. The height of the bell pepper plant showed a sigmoidal growth curve with a rapid vegetative growth from the transplanting until 65 days after transplantation (DAT). The maximum rate was of 1.1 $cmday^{-1}$ on open field in $I_{0.75}$ and under shade was 70 DAT, with a maximum of 1.32 $cmday^{-1}$ in $I_{0.50}$, according to the results of polynomial regression curve. Valles et al. (2009), found out that pepper plants showed a sigmoidal growth curve in where a rapid vegetative growth from the transplanting until 47 DAT, moment from which the

Table 3. Average of the absolute growth rate of the plant height and stem diameter of the bell pepper in open field and under shade mesh.

Treatment	Open field		Shade mesh	
	Plant height (cm day^{-1})	Stem diameter (mm day^{-1})	Plant height (cm day^{-1})	Stem diameter (mm day^{-1})
I_0	0.75	0.12^{b}	0,97	0.11^{b}
$I_{0.25}$	0.79	0.13^{ab}	1,01	0.13^{a}
$I_{0.50}$	0.85	0.13^{ab}	1.00	0.13^{a}
$I_{0.75}$	0.85	0.15^{a}	0.91	0.11^{ab}
$I_{1.0}$	0.81	0.14^{ab}	1.01	0.12^{ab}
Significance	ns	*	ns	*

Letters indicate significant differences at $^{*}P<0.05$ and $^{**}P<0.01$.

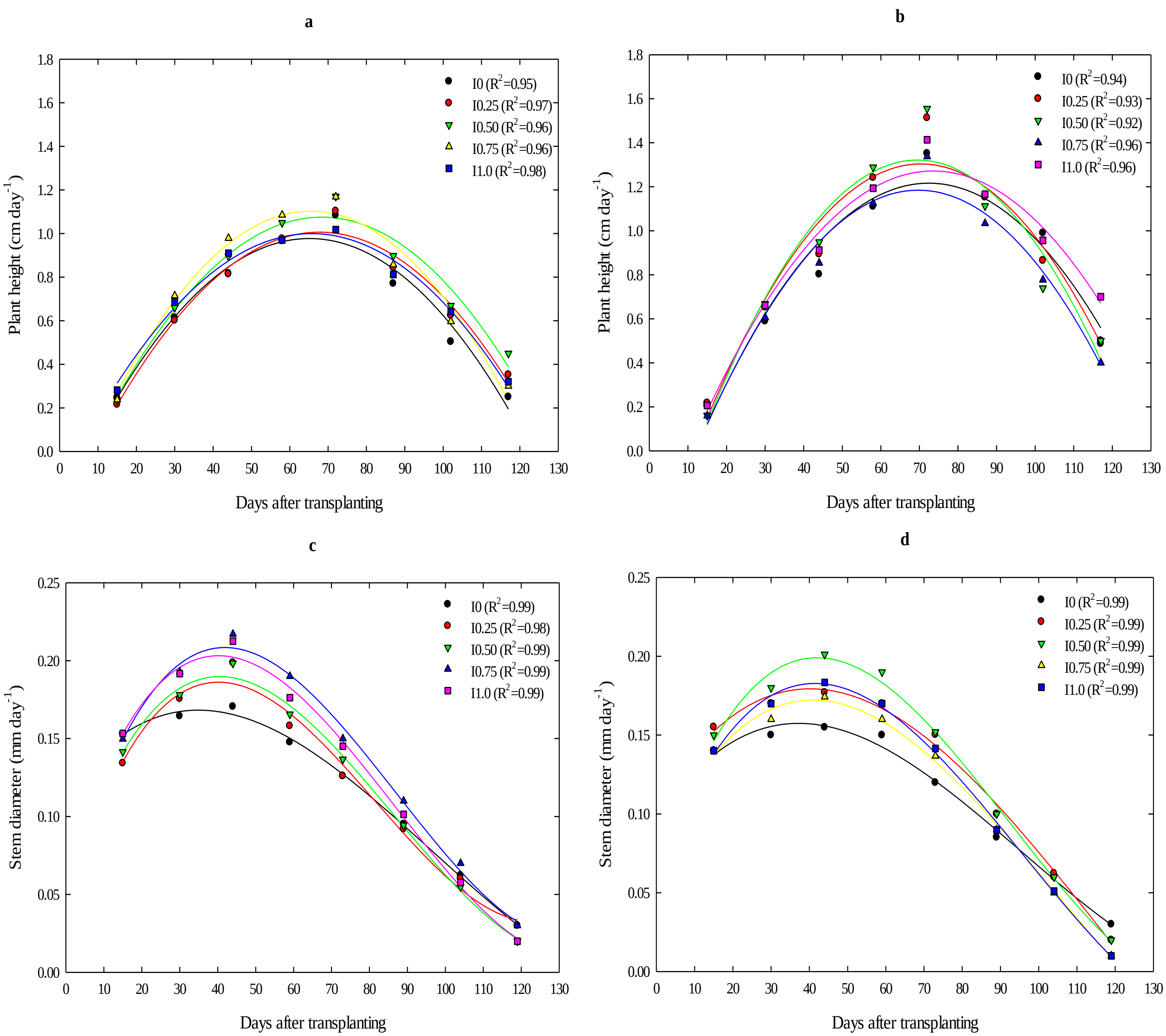

Figure 2. The absolute growth rates of the plant height and stem diameter of bell pepper in open field (a, c) and under shade mesh (b, d).

Table 4. Average leaf area index and number of leaves per plant at 90 days after transplanting of the bell pepper in open field and under shade mesh.

Treatment	Open field		Shade mesh	
	Leaf area index	leaves per plant (number)	Leaf area index	leaves per plant (number)
I_0	0.43^{cB}	119^{bB}	0.95^{bA}	187^{abA}
$I_{0.25}$	0.60^{bcB}	131^{abB}	0.95^{bA}	215^{bA}
$I_{0.50}$	1.01^{aA}	204^{aA}	1.13^{bA}	209^{aBA}
$I_{0.75}$	0.73^{bB}	144^{abB}	1.51^{aA}	269^{abA}
$I_{1.0}$	0.45^{cB}	121^{bB}	1.48^{aA}	316^{aA}
Significance	*	*	*	*

Letters indicate significant differences at $^{*}P<0.05$ and $^{**}P<0.01$.

growth rhythm decreased toward 62 DAT. This change point in growth rate corresponded with the formation of reproductive structures, which confirmed the undetermined growth of pepper plant.

The stem diameter showed rapid growth until 40 DAT in $I_{0.75}$ and 43 DAT in $I_{0.50}$ on open field and under shade, respectively. Under shade the growth rate is slower compared to open field conditions. Generally, in open field plots, lower plant height and higher stem diameter was observed. These results are similar to Díaz-Pérez (2013), that studied shade net levels in bell pepper, reporting that the stem diameter and the plant height were increased with increasinglevels of shade.Stems under shade were thinner and presumably less lignified than those in higher light conditions. Also, Ayala et al. (2015), studying different shade mesh colors in bell pepper, reported that shading provide an increase in plant height.This effect occurs significantly with black mesh, beige, red and green, where the plants grew from 23.1 to 33.0% more than those grown in the open field. Similarly, Márquez et al. (2014), studied different shade mesh colors (30% shade) in the cultivation of cherry tomato andthey recorded an increase in final plant height in all treatments compared to open field. The increase in plant height is a response of the reduced light. In this regard, Rylski and Spigelman (1986), studied the effect of different levels of shading (0, 12, 26 and 47% shading) in the development of sweet pepper plants cultivar 'Maor', reported that plant height increased as light intensity decreased (29.9, 30.3, 35.9 and 40.2 cm), respectively. The increase in plant height of shaded plants was a result of both internode elongation and node number, the apical growth was strongest under the lowest radiation.

The average leaf area index and number of leaves per plant at 90 DAT, is shown in (Table 4). Treatments in open field and under shade mesh had significant interaction effect on the leaf area index and number of leaves per plant. In all plots under shade it could be seen a greater leaf area index and number of leaves per plant. It the open-field plots, $I_{0.50}$ and $I_{0.75}$, had a greater development during the crop cycle and under shade, $I_{1.0}$ and $I_{0.75}$ (Figure 3). In I_0 and $I_{1.0}$open field plots,a less leaf area index was observed and under shade, I_0 and $I_{0.25}$. The leaf area index of $I_{0.50}$ was similar in both environments. It could be infer that the leaf area index was affected by the deficit and excess water. Generally, excess damp have an adverse effect on treatment with 50% shading, possibly optimal humidity is 50% of ET. Yildirim et al. (2012), observed that the plant development parameters of the bell peppers, such as plant weight, canopy and stem diameter, and LAI, decreased according to the amount of water applied from 367 to 164 mm. Good plant development in terms of whole plant weight, leaf area and LAI were observed in the full irrigation treatment, and those parameters were, 340 gplant^{-1}, 4012.9 cm^2 and 1.22. Excess water application did not increase the quality and development parameters of the peppers. Rylski and Spigelman(1986), reported leaf size increased as light intensity decreased.In shaded environment, leaves were bigger, total leaf area measured between the first and the fourth flower node was about 60% greater than that on plants grown in full light. These results are similar with studies showing that pepper under shade has large internodes, larger leaves, greater leaf area, and thinner leaves (Kittas et al., 2008; Ayala et al., 2011; Díaz-Pérez, 2013).

Chlorophyll index was higher in open plots in all measurements and irrigation shows no significant effect (Figure 4). However, in open field plots, it was observed a significant effect (P<0.05) at 60 DAT, being I_0 the lowest. On the other hand, significant interaction between treatments in open field and under shade mesh was observed in 90 and 105 DAT. Díaz-Pérez (2013), studyingshade levels in bell pepper, reported that chlorophyll index decreases as shading levels increase. Also they noted that a possible cause of inaccuracy under shade is related to the increased thickness of the leaf. Xiao et al. (2012), studying the effect of low light on the characteristics of photosynthesis and chlorophyll as fluorescence during leaf area development of sweet pepper, reported that under low light chlorophyll content, net photosynthetic rate, photosynthetic apparent quantum

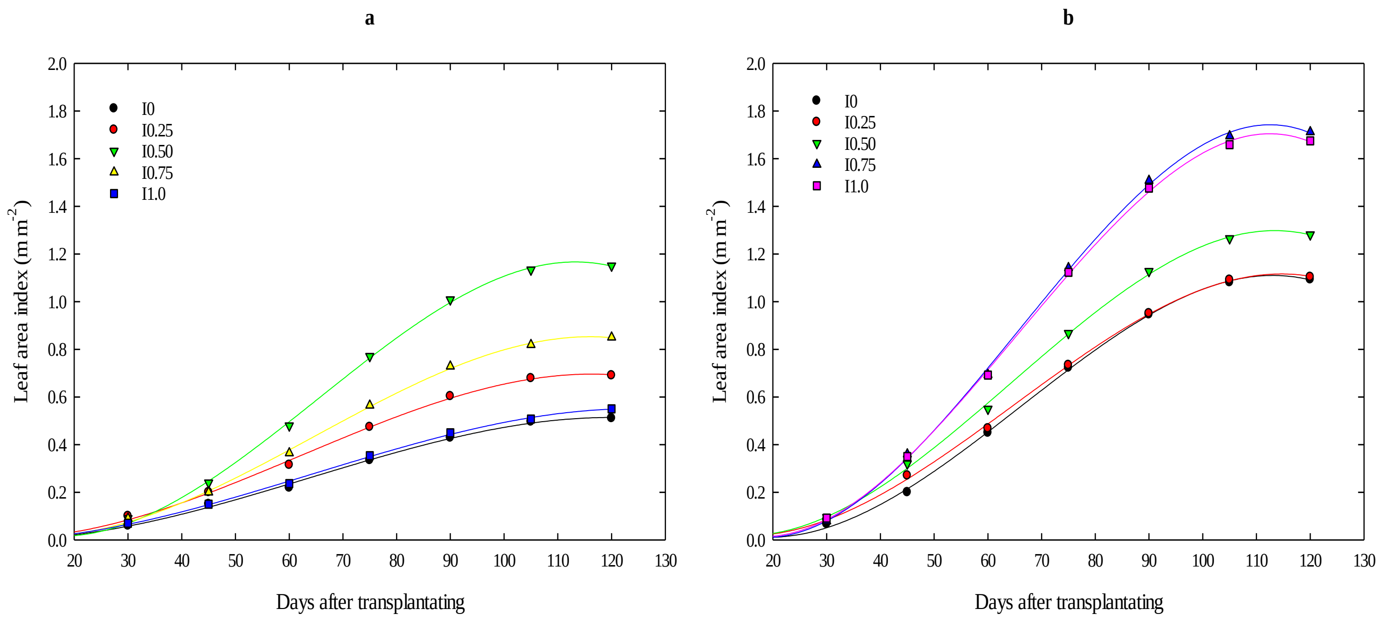

Figure 3. Leaf area index of the bell pepper in open field (a) and under shade mesh (b).

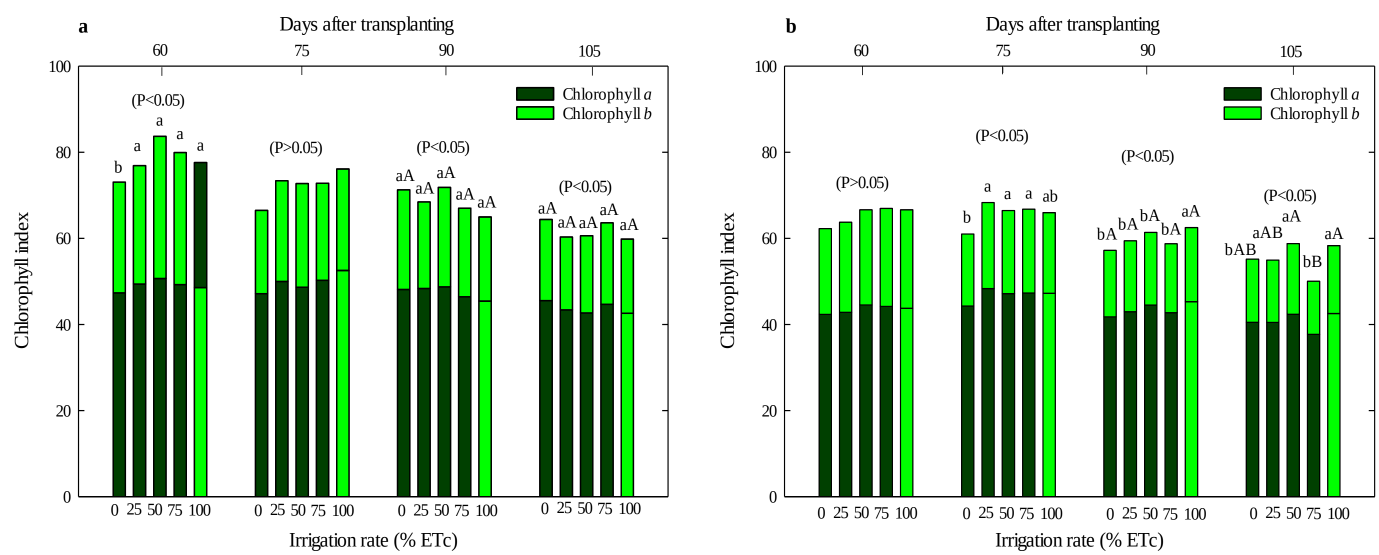

Figure 4. Chlorophyll content (index), Chlorophyll *a*, Chlorophyll *b* and Chlorophyll total of the bell pepper in open field (a) and under shade mesh (b).

efficiency and carboxylation efficiency of sweet pepper leaves increased gradually and decreased after reaching the maximum levels on 21st day under optimal light and the 42nd day under low light. Ferreyra et al. (1985), study chlorophyll content in pepper with different soil moistures, reported that with increasing soil moisture, apparently chlorophyll content decreases.

The average stem weight, root and part vegetative top, is shown in (Table 5). The treatments in open field and under shade showed significant effect interaction on stem weight, root and part vegetative top (P<0.05). The treatments I0.75 and I1.0 showed greater weight on part vegetative top in open field and under shade on I0.50 and I1.0. This result correlates with the leaf area index. Possibly open field plants are more lignified. Root development was higher in the treatments I0.75 and I1.0

Table 5. Average of dry mass of stem, root and vegetative top of the bell pepper in open field and under shade mesh.

Treatment	Open field (g plant^{-1})			Shade mesh (g plant^{-1})		
	Vegetative top	Root and stem	Total	Vegetative top	Root and stem	Total
I_0	117.7^{eB}	38.7^{cB}	156.3	157.8^{dA}	44.6^{bB}	202.4
$I_{0.25}$	141.3^{dB}	39.8^{cA}	181.1	150.5^{eA}	35.1^{dB}	185.6
$I_{0.50}$	145.1^{cB}	34.3^{dB}	179.4	228.4^{aA}	61.7^{aA}	290.1
$I_{0.75}$	206.7^{aA}	55.4^{bA}	262.0	164.2^{cB}	45.6^{bA}	209.7
$I_{1.0}$	201.7^{bA}	58.6^{aA}	260.3	179.9^{bB}	41.9^{cB}	221.8
Sig.	*	*	-	*	*	-

Letters indicate significant differences at $^{*}P<0.05$ and $^{**}P<0.01$. Sig: Significance.

in open field and under shade mesh on I0.50 and I0.75. This is due to excess moisture under shade, affecting soil aeration and consequently affecting root growth. In open field plots, weight of root and stem represent 21.9% and the aerial part of the plant 78.1%. The plant dry matter was 32.4 and 29.1%, in open field and under shade mesh, respectively.

The higher moisture content in soil was determined under shade mesh. Root development was mainly on 0 to 30 cm depth. Under shade, plants grew vigorously, leading to increased water consumption. On open field conditions, water consumption was lower, which contributed to low plant height and leaf area. Ferreyra et al. (1985), studied the effect of excessive use of water in pepper, reported that the total amount of root decreased markedly with excessive water application. Padrón et al. (2015b), studied irrigation levels in bell pepper, and reported that a decrease in irrigation water of 60% ET, roots grown deeper and the adventitious roots are thicker. On 100% of ET, ithas the highest vegetative growth. Nilsen and Orcutt (1996), reported that plants, as a strategy to increase water absorption capacity, increases the root surface, decreasing hydraulic resistance. This is common in plants known as wasteful water. Tambussi (2004), studying water saving plants, adopted as reverse strategy, minimizing water loss through various pathways, as: stomata closing and reducing perspiration coticule, within this same line conservative procedure. It could be included plants that produce less biomass to suffer water stress and increasing the relative proportion of the radicular mass.

Conclusions

The deficit and excess water affect on crop vegetative growth parameters. The determination of irrigation optimum levels provides vegetative and sustainable production characteristics. The cultivation of bell pepper with daily irrigation interval in a condition of 50% shading is recommended with the application of 0.50 of ETc and in open field 0.75 of ETc.These levels are not affected by the vegetative parameters of the crop.

Conflict of Interest

The authors have not declared any conflict of interest.

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