THE ROLE OF LIGHT REGIME AND SUBSTRATE IN PHOTOSYNTHETIC PIGMENTS, FREE PROLINE CONTENT AND FLOWER QUALITY OF GERBERA JAMESONII L.

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THE ROLE OF LIGHT REGIME AND SUBSTRATE IN PHOTOSYNTHETIC PIGMENTS, FREE PROLINE CONTENT AND FLOWER QUALITY OF *GERBERA JAMESONII* L.

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SUMMARY

The research of light and substrate role in leaf photosynthetic pigments, free proline content and flower quality indices in two gerbera cultivars (Vino and Ruby Red) showed that flower stems were longer in additional lighting treatment, coco's peat amended with rice chaff and cultivar Ruby Red. Chlorophyll b concentration was in negative relation to leaf proline content and correlated to longer flower stems. In general, the higher leaf total chlorophyll concentration was related to longer stems, and such stems have had larger flower heads. Flower head diameter did not differ significantly between cultivars; it depended on light regime, substrate and their interaction.

***Key-words**: light regime, substrate, photosynthetic pigments, proline, flower quality, gerbera*

INTRODUCTION

The economic importance of the ornamental plant production has shown remarkable growth recently, as stated by Horváth et al. (2006). Gerbera offers great potential for comparative developmental research within a single genotype, whereas different gerbera varieties show an impressive spectrum of color patterns, directly displaying responses to developmental cues at all important morphological levels (Teeri et al., 2006).
Supplementary lighting is a key factor in year-round gerbera growing in poor natural light conditions, so both the net growth and flower yield would be extremely limited during the darkest winter periods (Autio, 2000). The results from in vivo and in vitro experiments in gerbera (Meng et al., 2004) provide additional evidence that light is one of the most important factors for inducing flower pigmentation. Low light causes decreases in growth and photosynthesis of higher plants, that may result in the possibility of using photosynthetic parameters in low light tolerance screening (Sevelius, 2003; Lepeduš et al., 2005). On the other hand, as stated by Virupakshi et al. (2002), the development of inflorescence in sugarcane may be due to the photoperiod response or the use of proline in the culture medium, or it may be due to the cumulative effect of the above. Moreover, it was suggested that proline plays a key role in Arabidopsis flower transition, bolting and inflorescence formation (Mattioli et al., 2008).
Considering gerbera production, the soilless culture system was more profitable than the soil culture in the research of Grafiadellis et al. (2000). Potting media used for gerberas should be loose and well drained with a high percentage of organic matter. Many growers use 50 to 80 percent peat with perlite, vermiculite, calcine clay, or course sand added for the remaining percentage (Kessler, 1999). Experimentation on the physical and chemical properties of the substrate, with respect to particular plant characteristics helps optimize productivity in intensive culture methods (Issa et al., 2001).

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The evaluations of possible substrate x light regime interactions in gerbera soilless production might contribute to the optimization of growth conditions for this ornamental crop. Therefore, the aim of this research was to evaluate the impact of different light regime and two potting substrate on photosynthetic pigments and free proline concentration in gerbera leaves, as well as on some flower quality indices in two gerbera cultivars grown in a glasshouse in eastern Croatia climate.

MATERIAL AND METHODS

This research was designed as split-split-plot experiment with the factors as follows: light regime (A), substrate (B) and gerbera cultivar (C). Gerbera varieties used in this research were highly commercial Vino (C1) and Ruby Red (C2). They were planted (one plant per 3 L pot) in fall of 2005 in glasshouse of D.G. Promet in Magadenovac, East Croatia. Two types of substrate were used at planting, a pure cocos peat (B1) and cocos peat amended with rice chaff (40%; B2). Plants were fertilized with computer-based nutrient solution dosage depending on sun radiation intensity. The nutrient solution was composed of NO_3^- (11.25 mM dm^{-3}), NH_4^+ (1.5 mM dm^{-3}), P (1.25 mM dm^{-3}), S (1.25 mM dm^{-3}), K (5.5 mM dm^{-3}), Ca (3.0 mM dm^{-3}), Mg (1.0 mM dm^{-3}), Fe (35 μM dm^{-3}), Mn (5.0 mM dm^{-3}), Zn (4.0 μM dm^{-3}), B (30 μM dm^{-3}), Cu (0.75 μM dm^{-3}) and Mo (0.50 μM dm^{-3}). Solution pH was adjusted to 5.5 and EC was 1.5-2.0 mS cm^{-1}. Two levels of irradiance were applied: natural light during the whole growth period (variant A1) and additional lighting using HID lamps of 600 W ("High Intensity Discharge", variant A2), in the period November 11, 2006 to April 4, 2007 at the experimental glasshouse area having in total 320 plants. Each lamp supplied 20 plants (two repetitions) giving 9600 lux at 1.80 m, at flower heads level, and 5 600 lux at flower bud level, respectively. The artificial irradiance was applied daily between 9 h AM to 9 h PM. One week after the start of an additional light treatment and three times thereafter until the end of experimental period, plant leaves were collected for photosynthetic pigments and free proline content analyses. The individual levels of chlorophyll *a* (Chl a), chlorophyll *b* (Chl b), total chlorophylls (Chl) and total carotenoids (Car) in leaves were determined from acetone extract after Wettstein (1957) and expressed as mg g^{-1} leaf fresh mass. Free proline concentration (PRO) in gerbera leaves (μM g^{-1} leaf fresh mass) was determined after Bates et al. (1973). Each sample consisted of 10 leaves per repetition. At the same leaf analysis interval, flowers were detached from plants and their flower stem length (S-L) as well as flower head diameter (F-D) were measured. Data processing and statistical analyses were conducted with Statistica 7.1 data analysis software system (StatSoft, Inc., Tulsa, USA, 2004.).

RESULTS AND DISCUSSION

Photoperiod has a qualitative or quantitative effect on flower induction and development of some plant species, and no effect on flowering of other plants (Runkle and Heins, 2006). Not only supplementary lighting treatments did significantly increase vegetative growth and enhance flowering of tested gerbera cultivars, but also shortened production time compared to plants grown under ambient light conditions (control) in the research of Gagnon and Dansereau (1990). Differences among treatments were greater during the fall-winter study than during the winter-spring period. In the research presented here, the enhanced illumination treatment of two potted gerbera cultivars grown in the glasshouse, increased photosynthetic pigments content in general and resulted with better flower quality as compared with gerbera plants grown in ambient light conditions.

The lowest Chl a concentration had A1B1C1 variant and the highest was detected in A2B1C2 and A2B2C2 (Table 1), as a result of very significant influence ($P \leq 0.01$) of light regime (A), cultivar (C) and their interaction (AxC), as well as significant interactions BxC and AxBxC ($P \leq 0.05$). Chl b concentration was influenced mostly by cultivar and its interactions with other factors. It was the lowest in A1B1C1, the trial variant that showed the lowest concentration of pigments and the smallest flowers. Chl were not influenced by substrate (B) and light x substrate interaction (AxB), while the other factors and interactions were very significant. No change in chlorophyll levels, either time-wise or between gerbera cultivars, was observed in response to different substrates in the research of Issa et al. (2001). Here, Chl concentration was generally higher in A2 treatment (supplemental lighting), and C2 (cv. Ruby Red) had higher Chl concentration than C1 (cv. Vino) regardless light regime (Table 2). Car were not influenced by substrate and AxBxC interaction. The Chl a/b ratio was influenced by light and it was higher in A2 treatment. On the contrary, Chl/Car ratio was influenced only by substrate x cultivar interaction (BxC).

Leaf PRO concentration was strongly influenced by substrate and cultivar as well as interaction AxBxC, showing higher level in plants grown in cocos peat amended with rice chaff (B2) and in cv. Vino (C1). The applied light treatments had no statisticaly significant impact on leaf proline accumulation in gerbera in our research. As stated by Ashraf and Foolad (2007), accumulation of proline under stress in many plant species has been correlated with stress tolerance, and its concentration has been shown to be generally higher in stress-tolerant plants. Based on the estimated pigments concentrations and flower quality, it can be assumed that PRO concentration in leaves was not related to plant stress in our research. Furthermore, Mattioli et al. (2008) investigated the possible role of proline in flowering of Arabidopsis, and altered the expression of *AtP5CS1*, encoding the rate-limiting enzyme of proline biosynthesis in plants. In their research, *35S-P5CS1* plants manifested the overexpression of *P5CS1* and accumulation of proline early in development, leading to early flowering, both under long- and short-day conditions.

Table 1. Mean values of photosyntetic pigments content (Chl – chlorophyll, Car – carotenoids), their ratios, proline concentration (PRO), gerbera flower stem length (S-L) and flower diameter (F-D), under influence of light regime (A1-natural light; A2-supplemental lighting) in a glasshouse, substrate (B1-cocos peat; B2-cocos peat 60% + rice chaff 40%) and cultivar (C1-Vino; C2-Ruby Red). Each data item represents four repetitions and four measurements (November – April)

Tablica 1. Srednje vrijednosti sadržaja fotosintetskih pigmenata (Chl – klorofil, Car – karotenoidi), njihov omjer, koncentracija prolina (PRO), dužina cvjetne stapke (S-L) i promjer cvijeta gerbera (F-D) pod utjecajem svjetlosnog režima (A1-prirodno svjetlo; A2-dodatno osvjetljenje) u stakleniku, substrata (B1-kokosov treset; B2-kokosov treset 60% + rižina ljuska 40%) i kultivara (C1-Vino; C2-Ruby Red). Svaki podatak predstavlja prosjek četiri ponavljanja i četiri mjerenja (studeni-travanj)

	Chl a	Chl b	Chl	Car	Chl a/b	Chl/Car	PRO	S-L	F-D
	mg g^{-1} leaf fresh mass *mg g^{-1} u svježoj masi lista*						µM g^{-1} leaf fresh mass *µM g^{-1} u svježoj masi lista*	cm *cm*	
A1B1C1	1.36	0.47	1.83	0.76	2.89	2.41	2.79	42.70	8.45
A1B1C2	1.78	0.65	2.42	0.99	2.74	2.44	2.47	45.69	8.56
A1B2C1	1.67	0.60	2.27	0.96	2.86	2.35	3.48	48.82	9.80
A1B2C2	1.79	0.62	2.41	0.98	2.89	2.49	2.58	46.98	8.92
A2B1C1	1.82	0.58	2.40	1.00	3.17	2.40	3.02	48.54	9.61
A2B1C2	1.87	0.63	2.50	1.15	3.01	2.19	2.37	55.06	10.13
A2B2C1	1.80	0.60	2.41	1.10	3.02	2.20	3.20	55.09	10.36
A2B2C2	1.87	0.64	2.51	1.04	2.97	2.43	3.19	54.52	10.58
Analyses of variance, F-test: * *P≤0.05*, ** *P≤0.01*, *NS - non significant* *Analiza varijance, F test:* * P≤0.05, ** P≤0.01, NS - nije značajno									
A	**	NS	**	**	*	NS	NS	**	**
B	NS	NS	NS	NS	NS	NS	**	**	**
C	**	**	**	**	NS	NS	**	NS	NS
AxB	NS	NS	NS	*	NS	NS	NS	NS	*
AxC	**	NS	**	*	NS	NS	NS	NS	**
BxC	*	*	**	**	NS	**	NS	**	**
AxBxC	*	*	**	NS	NS	NS	**	NS	*

Table 2. Treatment means differences in photosyntetic pigments content and ratios (Chl – chlorophyll, Car – carotenoids), leaf proline concentration (PRO), gerbera flower stem length (S-L) and flower diameter (F-D); light regime: A1-natural light; A2-supplemental lighting; substrate: B1-cocos peat; B2-cocos peat 60% + rice chaff 40%; cultivar: C1-Vino; C2-Ruby Red. $^{a, b}$ – significantly different after $LSD_{0,05}$; ^{A,B} - significantly different after $LSD_{0,01}$

Tablica 2. Srednje vrijednosti tretmana u sadržaju i omjerima fotosintetskih pigmenata (Chl – klorofil, Car – karotenoidi), koncentraciji prolina u listu (PRO), dužini cvjetne stapke (S-L) i promjeru cvijeta (F-D) kod gerbera; svjetlosni režim: A1-prirodno svjetlo; A2-dodatno osvjetljenje; substrat: B1-kokosov treset; B2-kokosov treset 60% + rižine ljuske 40%; kultivar: C1-Vino; C2-Ruby Red. $^{a, b}$ – značajno različito prema $LSD_{0,05}$; $^{A, B}$ – značajno različito prema $LSD_{0,01}$

	Chl a	Chl b	Chl	Car	Chl a/b	Chl/Car	PRO	S-L	F-D
	Mg g^{-1} leaf fresh mass *Mg g^{-1} u svježoj masi lista*						µM g^{-1} leaf fresh mass *µM g^{-1} u svježoj masi lista*	cm *cm*	
A1	1.65^{A}	0.58^{A}	2.23^{A}	0.92^{A}	2.85^{A}	2.42	2.83	46.06^{A}	8.93^{A}
A2	1.84^{B}	0.62^{B}	2.46^{B}	1.07^{B}	3.04^{B}	2.30	2.95	53.33^{B}	10.16^{B}
B1	1.71	0.58	2.29	0.98	2.95	2.36	2.66^{A}	48.01^{A}	9.18^{A}
B2	1.78	0.62	2.40	1.02	2.94	2.37	3.11^{B}	51.38^{B}	9.91^{B}
C1	1.66^{A}	0.56^{A}	2.23^{A}	0.96^{A}	2.99	2.34	3.12^{B}	48.80^{a}	9.54
C2	1.83^{B}	0.64^{B}	2.46^{B}	1.04^{B}	2.90	2.39	2.65^{A}	50.59^{b}	9.54

As reported by Pettersen and Gislerød (2003), in the first part of the experimental period when the natural light conditions were poor, plants grown under 20 h lighting period produced the highest yield. However, when the natural light conditions improved in the spring, the plants grown under 10 h lighting period, partly or continuously, produced the highest yield. Autio (2000) stated that if gerbera was grown in extremely low natural light conditions, the availability of photoassimilates must be ensured to prevent reduction in inflorescence quality. We observed longer flower stems in supplemental lighting treatment (A2), B2 (cocos peat + rice chaff) and C2 (cv. Ruby Red). F-D did not differ significantly between cultivars but it was significantly influenced by other factors and interactions. These results confirm positive effect of supplemental lighting in glasshouse production of gerbera during fall-winter period in eastern Croatia conditions, regarding inflorescence quality indices presented here. Tested cultivars differed in pigments and PRO concentration, but not in pigment ratios or flower quality indices.

Substrate type was not significant in the accumulation and ratios of photosynthetic pigments in gerbera leaves but influenced very significantly on PRO and flower quality ($P \leq 0.01$). The results of Pisanu et al. (1994) pointed out statistically significant differences in the gerbera productions obtained in the different substrates. These differences were less appreciable in autumn and winter, while they were more marked in the spring-summer period. Moreover, a positive interaction was found between variety and substrate in relation to the quality of the yields. The interaction of light regime and substrate (AxB) in our research was significant only for Car and F-D. Chl a and Chl b concentration as well as F-D were influenced by this interaction at $P \leq 0.05$. On the contrary, substrate and cultivar interaction (BxC) had an impact on the majority of tested parameters with the exception of leaf PRO content and Chl a/b ratio. The interaction of all three factors (AxBxC) was significant ($P \leq 0.05$) for Chl a, Chl b and F-D, and very significant ($P \leq 0.01$) for Chl and PRO.

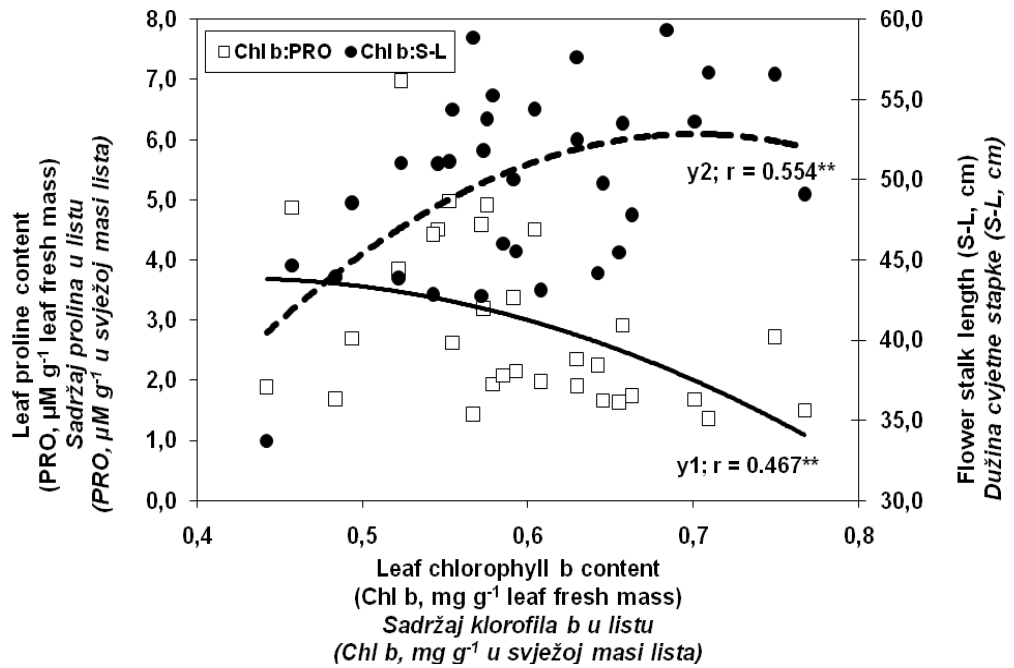

Figure 1. The relations among gerbera leaf Chl b (x axis), leaf proline content (left y axis, y1) and flower stem length (right y axis, y2), respectively

Grafikon 1. Veza između koncentracije Chl b (x os) u listu, sadržaja prolina u listu (lijeva y os, y1) i dužine cvjetna stapke (desna y os, y2) kod gerbera

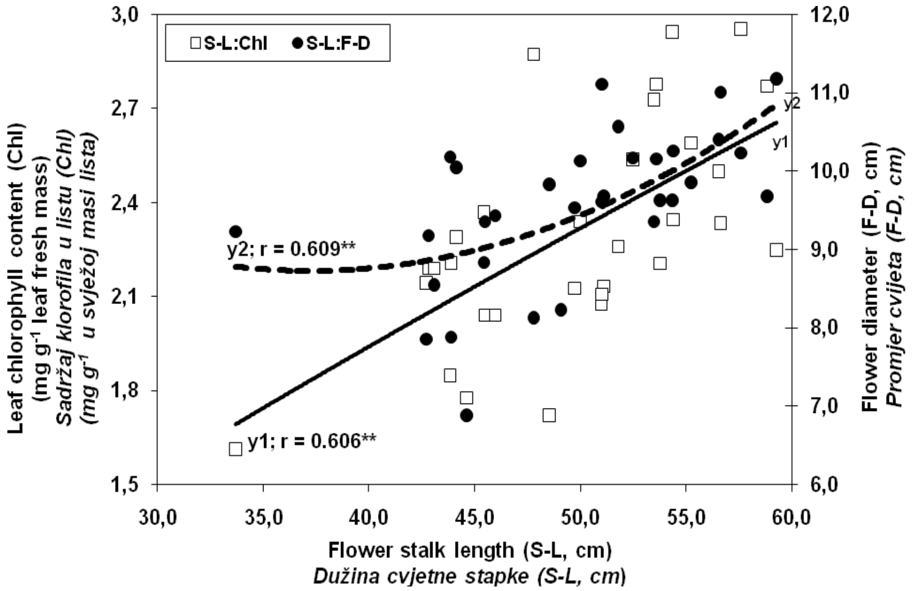

Figure 2. The relations among gerbera flower stem length (x axis), leaf Chl content (left y axis, y1) and flower head diameter (right y axis, y2), respectively

Grafikon 2. Veza između dužine cvjetne stapke (x os), koncentracije Chl u listu (lijeva y os, y1) i promjera cvjetne glavice (desna y os, y2) kod gerbera

Significant correlations among tested parameters showed that Chl b concentration was in negative relation to leaf PRO content (*Figure 1*; y1, r = 0.467**), and higher Chl b was correlated to longer flower stalks (*Figure 1*; y2, r = 0.554**). Consequently, higher Chl concentration was related to longer S-L (*Figure 2*, y1, r = 0.606**) and such flowers have had larger F-D (*Figure 2*, y2, r = 0.609**) in given experimental conditions. Multiple linear regression analyses performed with values taken as means of four determination in the experimental period, showed that F-D was significantly influenced by analyzed parameters ($F_{(8,32)}$= 23.87**, R^2 = 0.89**), with PRO and S-L having very significant impact (t – test values for Pro = 3.74** and for S-L = 7.09**).

CONCLUSION

This research showed that gerbera flower stems were longer in additional lighting treatment, cocos peat amended with rice chaff and cv. Ruby Red. Chl b concentration was in negative relation to leaf PRO content and correlated to longer F-S. In general, the higher leaf Chl concentration was related to longer F-S, and such flowers have had larger flower heads. F-D did not differ significantly between cultivars but it was dependent on light regime and substrate, as larger flower heads were observed in additional lighting treatment whereas cocos peat was amended with rice chaff. The interaction of light regime and substrate type was also statistically significant in the evaluated flower quality indices.

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ULOGA SVJETLOSNOGA REŽIMA I SUPSTRATA U SADRŽAJU FOTOSINTETSKIH PIGMENATA I SLOBODNOGA PROLINA TE KVALITETI CVIJETA KOD DVIJE SORTE GERBERA (*GERBERA JAMESONII L.*)

SAŽETAK

Istraživanje uloge svjetla i supstrata u sadržaju fotosintetskih pigmenata i slobodnoga prolina u listu, kao i pokazatelja kvalitete cvijeta kod dvije sorte gerbera (Vino i Ruby Red), pokazalo je da su cvjetne stapke bile duže u tretmanu s dodatnim osvjetljenjem, supstratu na bazi kokosa s dodanim rižinim pljevicama te kod sorte Ruby Red. Koncentracija klorofila b negativno je korelirala sa sadržajem prolina u listu i pozitivno s duljinom cvjetnih stapki. Općenito, viša koncentracija ukupnih klorofila bila je povezana s dužim stapkama i takve su stapke imale krupnije cvjetove. Promjer cvijeta nije se značajno razlikovao između sorata, a zavisio je o svjetlosnome režimu, supstratu i njihovoj interakciji.

Ključne riječi: gerber, karotenoidi, klorofil, prolin, svjetlosni režim, supstrat

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