ACTA AGROBOTANICA
Vol. 48, z. 2 – 1995
105-111

The influence of light quality on the shoot proliferation and rooting of *Gerbera jamesonii in vitro*

ELEONORA GABRYSZEWSKA, RYSZARD RUDNICKI

Research Institute of Pomology and Floriculture,
Pomologiczna 18, 96-100 Skierniewice, Poland

(Received: December 03, 1995)

Abstract

The effect of white, blue, green, red and UV + white light on the growth and development of shoots and roots of *Gerbera jamesonii* cv. Queen Rebecca in relation to the presence of kinetin or IAA were investigated. The highest number of axillary shoots was obtained in red and green light on the medium with 5 mg l^{-1} kinetin. Also, green and red light markedly increased the number of leaves developed on the plantlets on the medium supplemented with kinetin. Light quality and IAA added to culture medium variously affected the development of root system: roots were regenerated under all light treatments, higher root number was recorded under red light when 5 mg l^{-1} IAA was added to the media, the shortest roots were found in red light on the medium supplemented with IAA. The greatest fresh weight of shoots was found under white light on the medium with kinetin. Red light markedly decreased shoot fresh weight on hormone-free medium. Blue and white light caused increase in fresh weight of roots.

INTRODUCTION

Morphogenesis in plants *in vitro* is controlled by many factors: physiological stage of the explant donor, exogenous and endogenous hormones, organic and inorganic components and environmental factors (temperature, humidity and osmotic potential of the medium, gaseous phase and light).

Plant growth and development *in vitro* depend on light for photosynthesis and photomorphogenesis. Plant tissue cultures have been considered to have little photosynthetic ability and to require sugar as a source of carbon and energy for their heterotrophic or mixtrophic growth (K o z a i, 1991). Explants and plantlets *in vitro* require light mainly to regulate morphogenetic processes. Morphogenesis is influenced by hormones added to the medium and their endogenous levels in the culture shoots.

It was found that light can influence production and transport of hormones and can change sensitivity of tissue on hormones. Decreases in free IAA levels in

response to red and far-red light exposure have been demonstrated (I i n o, 1982). Also, light can induce changes in the levels of cytokinin-like substances (L e r c a r i and M i c h e l i, 1981) and can regulate transport of cytokinins (M a c h a c k o v a et al., 1995). Red/far-red light exposure combined with higher fluence exposure can induce the same degree of proliferation as that induced using higher exogenous BA levels (H e r r i n g t o n and M c P h e r s o n, 1993).

The effects of light quality on regeneration processes and shoot or root differentiation have been reported (B a r a l d i et al., 1988, 1994; F u e r n k r a n z et al., 1990; H e r r i n g t o n and M c P h e r s o n, 1993; M u l e o and M o r i n i, 1990; R o s s i et al., 1993; N o e et al., 1995).

This study aims to investigate the effect of light quality, and its possible interaction with kinetin or IAA, on growth and development of shoots and roots of *Gerbera jamesonii* cv. Queen Rebecca *in vitro*.

Abbreviations:
BA – 6-benzylaminopurine, 2iP – 6-(γ-γ dimethylallylamino)purine,
IAA – indole 3-acetic acid, MS – Murashige and Skoog medium,
UV – ultraviolet.

MATERIAL AND METHODS

Shoot culture of *Gerbera jamesonii* cv. Queen Rebecca were grown initially on a proliferation medium consisting of full strength MS salts (M u r a s h i g e and S k o o g, 1962), organic compounds and kinetin 5 mgl^{-1}. Shoots having at least 4-5 fully developed leaves were excised after 8 weeks of proliferation and were placed onto MS media containing 5 mgl^{-1} kinetin or 5 mgl^{-1} IAA and on the media without growth regulators. Cultures were incubated in growth chamber at 20-23°C under 16 h light /8 h dark cycles.

For experimental purposes the explants were maintained in different light quality (Tab. 1) under 16h light/8 h dark cycles.

T a b l e 1

Light characteristics in the experiment with gerbera cv. Queen Rebecca *in vitro*

Light quality	Tube type	Light intensity at culture level
White	Philips TLD 36W/95	88 $\mu mol \cdot m^{-2} \cdot s^{-1}$
UV + White	Philips TLD 36W/08 Philips TLD 36W/95	35 $\mu mol \cdot m^{-2} \cdot s^{-1}$
Blue	Philips TLD 36W/18	64 $\mu mol \cdot m^{-2} \cdot s^{-1}$
Green	Philips TLD 36W/17	30 $\mu mol \cdot m^{-2} \cdot s^{-1}$
Red	Philips TLD 36W/15	12 $\mu mol \cdot m^{-2} \cdot s^{-1}$

Twenty shoots were used per each treatment. After 8 weeks of culturing, the following parameters were determined:
– the number of shoots, leaves and roots per explant,
– the length of leaves and roots (mm),
– the fresh weight of shoots and roots (mg).

The results of experiment were interpreted statistically and subjected to analyses of variance. Duncan's test was used to estimate the difference between means at a 5 % level of significance.

RESULTS AND DISCUSSION

The mean numbers of shoots and leaves observed for the *in vitro* explants of gerbera under different light quality are represented in Tab. 2 and Tab. 3. Kinetin (5 mgl^{-1}) induced shoots and leaves formation in all light quality treatments; the highest numbers of axillary shoots and leaves were obtained under red and green light. There were not significant differences in number of leaves between light quality treatments in the absence of growth regulators or on the media containing IAA (5 mgl^{-1}). The presence of kinetin in the medium inhibited leaves elongation under all light treatments (Tab. 3). The longest leaves were observed under red, green and UV+white light in the absence of growth regulators or on the media supplemented with IAA (5 mgl^{-1}).

Roots were regenerated under all light treatments in the presence of IAA (5 mgl^{-1}) or kinetin (5 mgl^{-1}) and in the absence of growth regulators (Tab. 4). Red and green light markedly increased the number of formed roots if IAA (5 mgl^{-1}) was added to media. Also, the number of roots under blue light was greater than that under white light. In hormone-free medium the highest number of roots was found under white and blue light. Kinetin (5 mgl^{-1}) retarded the formation and growth of roots under all light quality treatments. The elongational growth of roots was strongly inhibited by red light in the presence of IAA (5 mgl^{-1}).

Table 2

The effect of light quality and kinetin 5 mgl^{-1} on the shoot proliferation of *Gerbera jamesonii* cv. Queen Rebecca *in vitro*

Light quality	Shoots number per plantlet
White	5.7 ab
UV + White	4.9 a
Blue	5.2 a
Green	7.2 bc
Red	7.7 c

Explanation: Means followed by the same letter are not significantly different (5 %) according to the Duncan's multiple range test.

Table 3

The effect of light quality, kinetin 5 mgl^{-1} and IAA 5 mgl^{-1} on the growth and development of leaves of *Gerbera jamesonii* cv. Queen Rebecca *in vitro*

Light quality	Growth regulators	Number of leaves per plantlet	Length of leaves (mm)
White	No growth regulators	8.9 a	44.5 cd
	Kinetin	23.6 b	21.4 a
	IAA	8.8 a	39.8 c
UV + White	No growth regulators	9.2 a	51.0 def
	Kinetin	21.8 b	29.0 b
	IAA	9.9 a	44.1 cd
Blue	No growth regulators	9.9 a	44.0 cd
	Kinetin	23.6 b	25.9 ab
	IAA	8.8 a	39.4 c
Green	No growth regulators	7.9 a	55.3 f
	Kinetin	32.6 c	24.8 ab
	IAA	10.7 a	47.6 de
Red	No growth regulators	8.8 a	50.5 def
	Kinetin	30.8 c	21.9 ab
	IAA	8.2 a	54.5 ef

Explanation: see Table 2

Table 4

The effect of light quality, IAA 5 mgl^{-1} and kinetin 5 mgl^{-1} on the growth and development of roots of *Gerbera jamesonii* cv. Queen Rebecca *in vitro*

Light quality	Growth regulators	Number of roots per plantlet	Length of roots (mm)
White	No growth regulators	5.6 ef	74.9 c
	IAA	6.4 fg	69.9 c
	Kinetin	2.3 abc	14.7 a
UV + White	No growth regulators	4.0 cde	80.7 c
	IAA	5.2 def	81.5 c
	Kinetin	0.8 a	25.1 ab
Blue	No growth regulators	5.1 ef	82.9 c
	IAA	7.9 g	77.5 c
	Kinetin	3.1 cd	26.1 ab
Green	No growth regulators	3.6 cde	71.6 c
	IAA	8.4 gh	65.6 c
	Kinetin	1.0 ab	15.6 a
Red	No growth regulators	3.0 bcd	69.8 c
	IAA	10.2 h	35.4 b
	Kinetin	1.0 ab	10.3 a

Explanation: see Table 2

Table 5

The effect of light quality, kinetin 5 mgl^{-1} and IAA 5 mgl^{-1} on the fresh weight of shoots and roots of *Gerbera jamesonii* cv. Queen Rebecca *in vitro*

Light quality	Growth regulators	Fresh weight of shoots (mg)	Fresh weight of roots (mg)
White	No growth regulators	569.5 abc	529.5 ef
	Kinetin	1005.4 e	72.3 ab
	IAA	522.6 ab	413.6 e
UV + White	No growth regulators	550.7 abc	282.6 d
	Kinetin	730.0 cd	31.7 ab
	IAA	555.0 abc	280.7 d
Blue	No growth regulators	653.6 bc	572.3 f
	Kinetin	838.7 de	152.7 bcd
	IAA	497.9 ab	455.1 ef
Green	No growth regulators	511.8 ab	253.6 d
	Kinetin	995.4e	25.1 a
	IAA	508.0 ab	93.4 ab
Red	No growth regulators	412.8 a	117.8 abc
	Kinetin	896.6 de	19.3 a
	IAA	550.5 abc	244.2 cd

Explanation: see Table 2

There were differences in shoot and root fresh weight among plants exposed to various light quality (Tab. 5). Shoots growing on the medium supplemented with kinetin produced the highest fresh weight under white light, whereas application UV + white light markedly decreased production of shoots fresh weight. Red light influenced lower production of shoot fresh weight values on hormone-free medium in comparison with other light treatments. Roots on plantlets growing under blue and white light, on the medium containing IAA or no hormones, produced the highest fresh weight. Kinetin in all light treatments significantly decreased fresh weight of roots.

Our results indicate the influence of spectral light quality and interaction between light and growth regulators in regulation of different types of organogenesis of *Gerbera jamesonii* cv. Queen Rebecca *in vitro*. Proliferation rate and number of leaves increased in red and green light only in the presence of 5 mgl^{-1} kinetin. Similar effects of red light were observed in regeneration of shoots in callus of *Actinidia deliciosa* on the medium with BA (M u l e o and M o r i n i, 1990). Also, shoots proliferation of *Ficus benjamina* under red light was promoted only in the presence of 2iP (G a b r y s z e w s k a and R u d n i c k i, in press). B a r a l d i et al. (1988) showed that the proliferation rate of *Prunus insititia* induced by BA under blue, far-red and white light strongly depended on the photon fluence rate, while no significant differences could be found under red irradiation at different photon fluence rate. It seems that the light-dependent BA induction of shoot formation can

be promoted by a low energy response of phytochrome. A red light may enable increased kinetin uptake into the explant from the growth medium and/or affect its subsequent metabolism. For example, the transport of cytokinins during the germination of chick-pea seeds is enhanced by red light (R e v i l l a et al., 1989).

Red and green light significantly stimulated rooting only in the presence of exogenously applied auxin (IAA). Red light was more effective in rhizogenesis than other light treatments. Similar effects of red light were observed by B a r a l d i et al. (1994) in root formation of *Pyrus communis* and by G a b r y s z e w s k a and R u d n i c k i (in press) in rooting of *Ficus benjamina* shoots in the presence of IAA. Red light in the absence or presence of auxin significantly enhanced rhizogenesis at low photon fluence rates, although further increase inhibited rooting (B a r a l d i et al., 1994).

In conclusion, the modification of endogenous hormones levels, their transport or sensitivity of tissue on endogenous or exogenous growth regulators, using light quality, may enable a reduction in exogenous growth regulators. Minimum required concentrations of exogenous growth regulators would provide advantages by reducing possible deleterious side-effects produced by high cytokinin and auxin levels.

Acknowledgements:
This work was support by Grant No. PBO 170/S3/93/04 from State Committee for Scientific Research (Poland).

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Wpływ jakości światła na proliferację i ukorzenianie pędów *Gerbera jamesonii in vitro*

S t r e s z c z e n i e

Badano wpływ światła białego, niebieskiego, czerwonego oraz UV + białego oraz współdziałanie jakości światła z kinetyną lub IAA we wzroście i rozwoju pędów oraz korzeni *Gerbera jamesonii* odm. Queen Rebecca *in vitro*. Najwięcej pędów kątowych powstawało w zespołach pędów namnażanych w świetle czerwonym i zielonym na pożywce zawierającej kinetynę. Także światło czerwone i zielone stymulowało tworzenie się liści u pędów rosnących na pożywce z kinetyną. Rodzaj stosowanego światła i obecność IAA w pożywce różnorodnie oddziaływały na powstawanie i rozwój korzeni na pędach: korzenie tworzyły się we wszystkich traktowaniach światłem; najwięcej korzeni stwierdzono na pędach ukorzenianych w świetle czerwonym na pożywce z IAA; także w tym traktowaniu obserwowano najkrótsze korzenie. Największą świeżą masę miały pędy namnażane w świetle białym, natomiast światło czerwone istotnie hamowało wzrost świeżej masy pędów na pożywce nie zawierającej regulatorów wzrostu. Światło niebieskie i białe wpływało korzystnie na wzrost świeżej masy korzeni.