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Prospective effects of methyl jasmonate to ameliorate *Celosia argentea* L. growth under low light conditions

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ABSTRACT

The current study was carried out to investigate the potential impact of methyl jasmonate (MeJA) at concentrations 0, 0.025, 0.05 and 0.1mM in raise Celosia argentea plant's ability to grow balanced under the influence of lack of light. When testing the plant for growth in a shaded place and comparing it with plants growing under sunlight, it was found that the shade treatment increased plant height, fresh and dry mass of spikes, photosynthetic pigments, flavonoids, and lipid peroxidation, whereas decreased the rest vegetative parameters (stem diameter, number of leaves/ plant, number of branches/ plant, leaf area, root length, fresh and dry mass of shoot and root), number of spikes/ plant, spike length, total sugars and anthocyanin content. The effect of MeJA on plants varied depending on light condition, where most vegetative parameters, all spike traits and anthocyanin pigment of the plants grown under full sunlight condition + 0mM MeJA produced the height values, while the treatment full sunlight+0.05 gave the highest values of chl.b, carotenoids, total sugars and flavonoids, but MDA was the highest with 0.1mM comparing with other treatment under the same light condition. On the other hand, the plants grown in shade were affected by MeJA treatment but the highest values for determined parameters fluctuated between concentrations 0.025 and 0.05mM. Based on the SDS-PAGE, the results showed that the plants treated with full sunlight + MeJA at 0, 0.05 and 0.1mM or shade + MeJA at 0 and 0.025 mM gave the largest number of polypeptides (9), while the lowest number of polypeptides (6) showed in the plants treated with shade+ MeJA at 0.1mM.

Keywords: Celosia argentea, methyl jasmonate (MeJA), shade, plant growth, lipid peroxidation, MDA, SDS-PAGE.

1. Introduction

Cockscomb (*Celosia argentea*) belongs the amaranth family (Amaranthaceae), is a grassy plant of tropical descent. It is a common to planted in the different garden. It produces dense undulating inflorescences that, hence it common name, the colors range from white to orange, red and purple. The spikes are used as cut flowers because they can hold for no more than 8 weeks and can be dried to use in floral arrangement. So, it has a considerable economic value as a cut flower and flowering pot (Surse *et al.*, 2014).

Among the major environmental agents, solar radiation is the most significant one that regulates the photosynthesis, and consequently, the plant survival, growth and adaptation but most of urban green spaces are located between the buildings, and 50% of them are found in shaded environments, which result in plants being exposed to limited sunlight and light intensity (Liu *et al.*, 2003). Consequently, decreasing in the biomass of plant as, photosynthetic rate, the transpiration and the stomatal conductance of water vapor (Wang *et al.*, 2009; Mielke and Schaffer, 2010).

Methyl jasmonate (MeJA) is a naturally occurring fragrant volatile ester form of jasmonic acid, exemplify one of the best known members of Jasmonates (JAs). It is known that MeJA has a

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substantial role in defence mechanisms to protect against both biotic and abiotic stresses (Cheong and Choi, 2003). methyl jasmonate is used to adjust responses through regulation of plant secondary metabolism (Meldau *et al.*, 2012), for example, by promoting the production of toxic compounds to protect and defence against herbivory (Moreira *et al.*, 2012) or by increasing reactive oxygen species (ROS) activity and concentration to protect against abiotic stresses (Anjum *et al.*, 2011). Because of the significant role of MeJA in the plant defence system, it has also been proposed that MeJA used as a chemical elicitor in young trees of conifer (Pinophyta) to raise their self-resistance ability, thereby increasing the survival rate of seedlings (Moreira *et al.*, 2012).

Del Amor and Cuadra-Crespo (2011); Wargent *et al.* (2013) reported that pre-treatment of plants with MeJA can alleviate the deleterious effects of salinity. However, it has also been stated that the exogenous application of MeJA can reduce photosynthesis (Attaran *et al.*, 2014) and prevent plant growth (Shyu and Brutnell, 2015). Till now it is considered that methyl esters of jasmonates (MeJA) as a chemical stress agent imitate the effect of that appear in response to external stress factors inducing symptoms of stress (Wasternack and Hause, 2002). MeJA inhibited chlorophyll accumulation at the level of chlorophyll precursors in the dark (Ananieva *et al.*, 2004) under different environmental conditions.

The ability of MeJA has been tested in many studies to help plants withstand different stress conditions, but there is a lack of studies that explain its effect in the absence of adequate light for plant growth, so the study goal was to bridge the gap at this point, and that is through clarification the extent of the negative impact of lack of lighting on the growth and development of the *C. argentea* plant, and to study the ability of methyl jasmonate to mitigate these damages and help the plant to resist and continue to grow under these conditions.

2. Materials and Methods

The experiment was conducted during seasons of 2021 and 2022 in the greenhouse of the Horticulture Research Institute (HRI), Agricultural Research Center (ARC). The seeds of *Celosia argentea* were obtained from HRI, ARC and sown in plastic trays on the mid of February during the two seasons; The early seedlings were transplanted on March when they started having 6 to 8 real leaves in 30cm pots filled with sand and clay, the physical and chemical analysis of the used media were shown in Table (1) which were determined according to George *et al.* (2013). The plants divided into two groups one of them was left to grow in the full sunlight and the other was placed under shade cloth (shading percentage 63%), both of the groups sprayed with the methyl Jasmonate (MeJA) at concentrations 0, 0.025, 0.05 and 0.1mM, all of the concentration applied twice where the first application was after one month from the transplanting and the second after one month from the first one. The light intensity for two groups was measured and recorded daily by device digital lux meter (BENETECH model: GM1020) and the average was calculated for each month during each season and inserted in Fig. (1).

The plants were collected in July for each season to record the vegetative and spikes measurements and chemical estimates which was conducted in the National Research Centre.

Tuble 1. Thysical and chemical analysis of the used media									
C	hemical analysis	F	Physical analysis						
рН	7.45	V.C.S	10.19%						
EC (1:1)	16.46 dS/m	C.S.	20.13%						
CO3 ²⁻	0.00	M.S.	41.25%						
HCO ₃ -	8.80 meq/ L	F.S.	14.41%						
Cl	112.0 meq/ L	V.F.S.	20.88%						
SO 4 ²⁻	34.66 meq/ L	Silt+Clay	6.21%						
Ca ²⁺	27.18 meq/ L								
Mg ²⁺	23.36 meq/ L								
K ⁺	15.04 meq/ L								
Na ⁺	89.08 meq/ L								

Table 1: Physical and chemical analysis of the used media

Where; V.C.S: very coarse sand, C.S.: coarse sand, M.S.: medium sand, F.S.: fine sand and V.F.S.: very fine sand



Fig. 1: The average of light intensity (lux) for the sunlight and shade 63% during 2021 and 2022 seasons

2.1. Vegetative measurements: plant height (cm), stem diameter (cm), number of leaves/ plant, number of branches/ plant, leaf area (cm²), root length (cm), fresh and dry mass of shoot and root (g).

2.2. Spike measurements: number of spikes/ plant, spike length (cm), fresh and dry mass of spikes.

2.3. Chemical estimates:

Photosynthetic pigments including chlorophyll a, b and carotenoids (mg. g^{-1} F.W.) were determined according to Saric *et al.* (1967).

Total sugars content (mg. g⁻¹ F.W.) was determined according to Dubois et al. (1956).

Total flavonoids content (µg. g⁻¹ F.W.) was determined according to Quettier et al. (2000).

Lipid peroxidation (μ g. g⁻¹ F.W.) was expressed as malondialdehyde (MDA) content and was determined according to Buege and Aust (1972).

Anthocyanin pigment in spikes was determined according to Husia et al. (1965).

2.4. Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE)

Young fresh leaves were collected from each treatments under investigation and were ground into a fine powder by using liquid nitrogen (-196 °C) and a mortar and pestle according to the method of Laemmli (1970) and modified by Studier (1973). The molecular weights of proteins were estimated relative to a standard protein marker with a wide range of molecular weight. Gel was digitally photographed and analyzed using Gel Doc Viller Lourmat system to capture the image and to calculate monomorphic and polymorphic bands.

2.5. Experiment layout and statistical analysis

The experimental layout was set in factorial experiment in complete block design with two irrigation intervals and sprayed with 9 rates of carrot and/ or turmeric extracts to give 18 treatments with 3 replicates for each season. The obtained results were subjected to statistical analysis by using least significant differences (LSD) at 5% level according to method described by Snedecor and Cochran (1980).

3. Results

3.1. Vegetative measurements

The data inserted in Tables (2&3) cleared that *C. argentea* plants exposed to full sunlight or shade conditions produced differences in the values of the measured traits whether morphological, flowering, chemical composition or pigments. Exposure of plants to shade led to an increase in plant

height (cm), giving average values of 27.31 and 28.58 followed by plants that were exposed to full sunlight, giving average values 27.18 and 27.77, respectively, in both seasons without significant differences between the both treatments. While the study showed that the rest of the vegetative traits are included stem diameter (cm), number of leaves/ plant, number of branches/ plant, leaf area (cm²), root length (cm), shoot fresh and dry mass (g) and root fresh and dry mass (g) giving values 0.85, 72.58, 15.75, 11.51, 16.65, 19.68, 2.03, 4.06 and 1.17, respectively, in the first season and 0.88, 79.25, 16.42, 10.30, 16.87, 19.68, 2.27, 4.20 and 1.20, respectively, in the second season.

 Table 2: Effect of different light condition (full sunlight or shade) and methyl jasmonate (MeJA) on plant height, stem diameter, no. of leaves/ plant, no. of branches/ plant and leaf area of *Celosia argentea* plant during 2021 and 2022 seasons

Treatment		Plant height (cm)		Stem diameter (cm)		No. of leaves/ plant		No. of branches/ plant		Leaf area (cm ²)	
		1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
		season	season	season	season	season	season	season	season	season	season
Full sun		27.18	27.77	0.85	0.88	72.58	79.25	15.75	16.42	11.51	10.30
Shade		27.31	28.58	0.77	0.78	63.17	71.84	12.75	13.92	8.92	8.28
LSD _{5%}		1.34	1.75	0.03	0.04	1.85	2.92	1.70	1.84	0.72	0.65
MeJA 0 m	ηΜ	25.30	26.45	0.83	0.86	41.17	49.34	9.50	9.84	12.12	11.21
MeJA 0.02	25mM	28.82	28.78	0.81	0.82	71.84	82.00	15.67	17.17	11.37	10.18
MeJA 0.05 mM		29.29	31.27	0.85	0.87	103.84	110.00	20.17	21.5	9.42	8.53
MeJA 0.1	mM	25.57	26.20	0.76	0.78	54.67	60.84	11.67	12.17	7.96	7.24
LSD _{5%}		1.90	2.47	0.04	0.06	2.62	4.13	2.41	2.60	1.02	0.91
	MeJA 0 mM	30.50	34.67	1.01	1.06	52.00	58.00	12.67	12.67	15.62	14.34
E-II ann	MeJA 0.025mM	27.47	30.03	0.89	0.92	78.00	86.33	16.33	17.67	11.79	9.96
r un sun	MeJA 0.05 mM	26.07	28.43	0.80	0.84	97.00	102.67	19.67	20.33	10.09	9.27
	MeJA 0.1 mM	24.67	25.73	0.68	0.70	63.33	70.00	14.33	15.00	8.55	7.64
	MeJA 0 mM	20.10	22.23	0.64	0.65	30.33	40.67	6.33	7.00	8.62	8.08
61 1	MeJA 0.025mM	30.17	29.52	0.73	0.71	65.67	77.67	15.00	16.67	10.95	10.4
Shade	MeJA 0.05 mM	32.50	35.90	0.89	0.90	110.67	117.33	20.67	22.67	8.74	7.79
	MeJA 0.1 mM	26.47	26.67	0.83	0.85	46.00	51.67	9.00	9.33	7.36	6.84
LSD _{5%}		2.68	3.49	0.05	0.08	3.71	5.84	3.41	4.51	1.45	1.29

 Table 3: Effect of different light condition (full sunlight or shade) and methyl jasmonate (MeJA) on root length, fresh and dry mass of shoot and root of *Celosia argentea* plant during 2021 and 2022 seasons

Treatment		Root length		Fresh	Fresh mass of		Dry mass of		Fresh mass of		nass of
		(C	(CIII)		<u>ρι (g)</u>	1st 2nd		1 st and			
		1	2	1	2	1	2	1	2	1	2
E 11		season	season	season	season	season	season	season	season	season	season
Full sun		16.65	16.87	19.68	19.68	2.03	2.27	4.06	4.20	1.17	1.20
Shade		14.71	14.77	16.95	18.30	1.65	1.82	3.12	3.08	0.88	0.85
LSD5%		1.25	1.18	0.90	0.96	0.25	0.16	0.51	0.40	0.10	0.10
MeJA 0 ml	М	16.19	15.87	19.71	19.90	2.09	2.36	4.10	4.05	1.19	1.15
MeJA 0.025mM		16.59	17.07	20.23	20.83	2.04	2.22	4.26	4.05	1.23	1.17
MeJA 0.05 mM		15.79	15.42	20.13	21.89	2.07	2.33	3.00	3.30	0.84	0.92
MeJA 0.1 mM		14.17	14.92	13.19	14.13	1.15	1.29	3.01	3.17	0.85	0.88
LSD _{5%}		1.76	1.66	1.27	1.35	0.36	0.22	0.72	0.56	0.13	0.14
	MeJA 0 mM	20.80	19.67	27.49	29.07	3.22	3.71	6.23	5.94	1.84	1.74
Eull ann	MeJA 0.025mM	15.10	15.80	21.88	22.68	2.29	2.61	3.65	3.73	1.04	1.06
r un sun	MeJA 0.05 mM	15.00	15.33	16.55	17.33	1.51	1.58	2.61	3.24	0.72	0.89
	MeJA 0.1 mM	15.70	16.67	12.80	14.02	1.08	1.18	3.75	3.90	1.08	1.12
	MeJA 0 mM	11.57	12.07	11.92	12.31	0.96	1.00	1.97	2.16	0.53	0.56
Shada	MeJA 0.025mM	18.07	18.33	18.58	18.97	1.79	1.83	4.87	4.37	1.41	1.27
Shaue	MeJA 0.05 mM	16.57	15.50	23.71	26.44	2.62	3.07	3.38	3.35	0.95	0.94
	MeJA 0.1 mM	12.63	13.17	13.57	15.46	1.21	1.39	2.26	2.44	0.61	0.64
LSD _{5%}		2.49	2.35	1.79	1.91	0.51	0.31	1.02	0.80	0.19	0.20

In the same Tables mentioned in the previous paragraph, the results showed that MeJA treatment had a clear effect on plant characteristics, which differed from one concentration to another. Where, the treatment with concentration 0.05 mM gave the highest increase in the values of plant height (29.29 and 31.27), stem diameter (0.85 and 0.87), number of leaves (103.84 and 110.00), number of branches (20.17 and 21.50), respectively, in both seasons, and fresh mass of shoots (21.89)

in the second season, while the plants that were exposed to the concentration of 0.025mM showed the highest increase in the values of root length (16.59 and 17.07), and dry mass of the root (1.23 and 1.17), respectively, in both seasons and the fresh mass of shoots in the first season (4.26), as for the fresh mass of the root in the first season it gave (4.26) while in the second season the untreated plants and plants treated with concentration 0.025mM gave the same value (4.05), the highest value of leaf area was found in control plants giving 12.12 and 11.21, respectively, in both seasons.

As for the effect of the interaction of MeJA on plants growing in full sunlight or shade conditions, the results showed that plants not treated with MeJA and exposed to full sunlight gave the highest values for all measured vegetative traits except for the number of leaves and the number of branches, which were their highest values are at a concentration of 0.05 mM. On the other hand, shade-exposed plants that were treated with MeJA at a concentration of 0.05 mM showed the highest values of plant height, stem diameter, number of branches, fresh and dry mass of shoots, while the rest of the traits were positively affected when treated at a concentration of 0.025 mM.

3.2. Spike traits

The symptoms of light variation appeared clearly on the characteristics of the inflorescences of the *C. argentea* plant, where the data attached in Table (4) showed that the highest values of number of spikes/ plant (11.92 and 13.67, respectively, in the 1st and 2nd seasons) and spike length (4.43 and 4.59 cm, respectively, in the 1st and 2nd seasons) were obtained from plants grown in full sunlight condition. Whereas, the highest values of fresh mass of spikes (4.85 and 5.28g, respectively, in the 1st and 2nd seasons) and dry mass of spikes (0.72 and 0.77g, respectively, in the 1st and 2nd seasons) was produced in plants grown in the shade conditions.

Treatment		No. of	No. of spikes/ plant		Spike length (cm)		Fresh mass of spike (g)		nass of e (g)
		1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
		season	season	season	season	season	season	season	season
Full sun		11.92	13.67	4.43	4.59	4.75	5.15	0.68	0.72
Shade		9.92	11.92	3.70	3.84	4.85	5.28	0.72	0.77
LSD _{5%}		0.92	1.31	0.48	0.40	0.39	0.30	0.05	0.04
MeJA 0 mM		8.67	9.67	4.44	4.53	4.1	4.01	0.62	0.58
MeJA 0.025mM		9.67	11.34	5.29	5.47	3.96	4.71	0.54	0.66
MeJA 0.05 mM		15.83	17.84	4.12	4.27	7.18	7.64	1.09	1.15
MeJA 0.1 mM	M	9.50	12.34	2.42	2.57	3.96	4.51	0.55	0.60
LSD _{5%}		1.32	1.85	0.67	0.56	0.55	0.40	0.07	0.06
	MeJA 0 mM	15.67	17.33	6.27	6.43	7.58	7.28	1.15	1.07
Full cun	MeJA 0.025mM	13.00	14.67	4.80	4.87	5.07	5.72	0.70	0.82
r un sun	MeJA 0.05 mM	11.33	12.00	3.97	4.17	3.97	4.20	0.54	0.57
	MeJA 0.1 mM	7.67	10.67	2.67	2.87	2.38	3.41	0.31	0.43
	MeJA 0 mM	1.67	2.00	2.60	2.63	0.62	0.74	0.08	0.09
Shada	MeJA 0.025mM	6.33	8.00	5.77	6.07	2.85	3.60	0.38	0.49
Shaue	MeJA 0.05 mM	20.33	23.67	4.27	4.37	10.38	11.07	1.63	1.73
	MeJA 0.1 mM	11.33	14.00	2.17	2.27	5.53	5.60	0.79	0.77
LSD _{5%}		1.86	2.62	0.95	0.80	0.77	0.56	0.10	0.09

 Table 4: Effect of different light condition (full sunlight or shade) and methyl jasmonate (MeJA) on spikes traits of *Celosia argentea* plant during 2021 and 2022 seasons

It was found that when calculating the mean average of the values of the spike traits that were estimated in the study that the treatment with MeJA showed significant differences in most of the traits mentioned, which were concluded through statistical analysis of the obtained values. The data recorded in Table (4) showed that application of MeJA at concentration of 0.05mM produced the highest values of number of spikes (15.83 and 17.84), fresh mass of spikes (7.18 and 7.64 g) and dry mass of spikes (1.09 and 1.15g), meanwhile the spike length was increased by application of MeJA at concentration of 0.025mM giving 5.29 and 5.47cm, respectively, in the first and second seasons as compared with control plants.

Regarding the interaction treatments, it was found that the plant grown in full sun light condition + 0 mM of MeJA gave the highest values of the measured traits and then decreased gradually by increasing the concentration of MeJA under the same light condition. While the shade-

exposed plants + 0.05 mM of MeJA gave the highest values of recorded traits of spike except spike length which increased with treatment shade + 0.025mM of MeJA in the first and second season.

3.3. Chemical determinations

3.3.1. Photosynthetic pigments and flavonoids

The growth of plants in shade conditions led to an increase in the leaves content of photosynthetic pigments and flavonoids, and this was evident in the data recorded in Table (5), where the values of chlorophyll a was 0.54 and 0.56, chlorophyll b was 0.18 and 0.19, carotenoids was 0.36 and 0.39 mg. g⁻¹ F.W. and flavonoids was 600.09 and 590.97 μ g. g⁻¹ F.W., respectively, in both seasons compared to plants grown under full sun.

The study showed that the treatment with MeJA led to an increase in the plant content of pigments, whether photosynthetic pigments or flavonoids. Spraying plants with MeJA at concentration 0.025mM increased the content of photosynthetic pigments giving values of 0.48, 0.16 and 0.33 in the first season and 0.48, 0.18 and 0.36 mg. g^{-1} F.W. in the second season for chlorophyll a, b and carotenoids, respectively. While the highest content of flavonoids (646.09 and 657.79 µg. g^{-1} F.W.) was given by the plants sprayed with concentration 0.05mM in both seasons, compared to the plants sprayed with distilled water.

While studying the effect of the interaction between plant growth in different lighting conditions and spraying with MeJA, it was found that in the case of plants growing under sun conditions, the plant content of chlorophyll a gradually increased with the increase in the concentration of MeJA, where the highest result was found in the treatment 0.1 mM while the lowest result was found in untreated plants, while the highest result of the remaining pigments (chlorophyll b, carotenoids and flavonoids) was obtained in plants treated with concentration 0.05 mM. Plants grown under shade conditions showed the highest content of all pigments when sprayed with concentration 0.025 compared to the rest of the treatments under the same light conditions.

3.3.2. Total sugars content

The results presented in the Table (5) showed that the total sugars content increased slightly, non-significantly, giving values 0.48 and 0.52 mg. g^{-1} F.W. in plants that were fully exposed to sunlight compared to plants grown in the shade giving values 0.45 and 0.49 mg. g^{-1} F.W., respectively, in both seasons.

Treating plants with MeJA led to a significant increase in plant content of total sugars compared to untreated plants, as the highest result was 0.53 mg. g^{-1} F.W. in the first season when treated with concentration 0.025, while in the second season, treatment with concentration 0.025 or 0.05 gave the highest result with the same the value 0.56 mg. g^{-1} F.W.

Plants grown in full sunlight and treated with MeJA at concentration 0.05mM, gave the highest values of 0.59 and 0.63 mg. g^{-1} F.W. As for plants grown in shade conditions, the highest values of sugar content appeared when treated with MeJA at concentration 0.025 mM giving values of 0.54 and 0.57 mg. g^{-1} F.W, compared to the rest treatments in both seasons.

3.3.3. Lipid peroxidation (MDA)

The data listed in Table (5) showed a significant increase in the content of plant leaves of MDA in plants grown in shade (0.79 and 0.75 μ g. g⁻¹ F.W.) compared to plants grown in sunlight (0.40 and 0.40 μ g. g⁻¹ F.W.) in both seasons.

Plants not treated with MeJA gave the highest content of MDA compared to the rest of the treatments, where the lowest content observed in plants that were sprayed with concentration 0.025mM, then it increased again gradually with increasing the concentration of MeJA, but it was lower than the control as well.

As for the interaction treatments, the plants grown in full sunlight and sprayed with distilled water gave the lowest content of MDA, but the content of MDA increased directly with the increase of the concentration of MeJA, as the highest content of MDA was found in plants that received MeJA at 0.1mM , while the plants which grown in the shade, its content of MDA was inversely with the increase in the concentration of MeJA, as the highest content was found in the untreated plants, while the lowest content was found in the plants treated with the concentration 0.1mM

Table 5: Effect of different	light condition (full s	sunlight or shade) and	l methyl jasmonate (MeJA) on
some chemical dete	erminations of Celosia	argentea plant durin	g 2021 and 2022 seasons

		Ch	l. a	Chl. b		Carotenoids		Total sugars	
Т	reatment	1 st	2 nd						
		season							
Full sun		0.32	0.34	0.11	0.12	0.21	0.23	0.48	0.52
Shade		0.54	0.56	0.18	0.19	0.36	0.39	0.45	0.49
LSD _{5%}		0.04	0.06	0.03	0.02	0.04	0.03	0.03	0.04
MeJA 0 m	Μ	0.42	0.42	0.14	0.14	0.26	0.29	0.44	0.48
MeJA 0.02	25mM	0.48	0.48	0.16	0.18	0.33	0.36	0.53	0.56
MeJA 0.05	5 mM	0.37	0.46	0.14	0.15	0.29	0.32	0.52	0.56
MeJA 0.1	mМ	0.44	0.45	0.14	0.15	0.28	0.28	0.37	0.42
LSD5%		0.05	0.08	0.05	0.03	0.05	0.04	0.04	0.05
	MeJA 0 mM	0.27	0.30	0.09	0.10	0.18	0.19	0.43	0.47
Eull cun	MeJA 0.025mM	0.30	0.32	0.10	0.12	0.21	0.23	0.51	0.55
r un sun	MeJA 0.05 mM	0.32	0.36	0.13	0.14	0.27	0.30	0.59	0.63
	MeJA 0.1 mM	0.37	0.39	0.10	0.11	0.19	0.20	0.38	0.42
	MeJA 0 mM	0.56	0.53	0.18	0.18	0.34	0.38	0.45	0.49
Shada	MeJA 0.025mM	0.65	0.64	0.21	0.23	0.44	0.48	0.54	0.57
Snade	MeJA 0.05 mM	0.42	0.56	0.15	0.16	0.31	0.33	0.44	0.49
	MeJA 0.1 mM	0.51	0.50	0.17	0.19	0.36	0.35	0.36	0.41
LSD5%		0.07	0.11	0.07	0.05	0.07	0.06	0.06	0.08

Table 5: Cont.

-		Flavo	onoids	M	DA	Anthocyanin		
	Treatment	1 st	2 nd	1 st	2 nd	1 st	2 nd	
		season	season	season	season	season	season	
Full sun		518.79	528.81	0.40	0.40	2.02	2.11	
Shade		600.09	590.97	0.79	0.75	1.94	2.00	
LSD5%		10.91	7.52	0.04	0.03	0.10	0.09	
MeJA 0 m	M	468.96	454.64	0.93	0.88	2.60	2.67	
MeJA 0.02	25mM	644.52	643.36	0.4	0.38	1.89	1.97	
MeJA 0.05	5 mM	646.09	657.79	0.43	0.45	1.73	1.83	
MeJA 0.1	mM	478.20	483.78	0.64	0.59	1.71	1.74	
LSD5%		15.43	10.63	0.05	0.04	0.15	0.12	
	MeJA 0 mM	433.74	423.27	0.10	0.09	2.71	2.78	
E-II and	MeJA 0.025mM	527.39	543.82	0.12	0.14	1.90	2.01	
Full Sun	MeJA 0.05 mM	677.42	688.90	0.35	0.38	1.74	1.86	
	MeJA 0.1 mM	436.61	459.25	1.04	0.97	1.73	1.77	
	MeJA 0 mM	504.18	486.01	1.76	1.66	2.48	2.56	
Shada	MeJA 0.025mM	761.65	742.89	0.68	0.62	1.87	1.93	
Shaue	MeJA 0.05 mM	614.75	626.68	0.50	0.51	1.71	1.79	
	MeJA 0.1 mM	519.79	508.30	0.23	0.20	1.68	1.70	
LSD5%		21.81	15.03	0.07	0.06	0.21	0.17	

3.3.4. Anthocyanin content in spikes

From the data recorded in Table (5), it was found that the highest content of anthocyanin pigment present in the inflorescences of the *C. argentea* plant (2.02 and 2.11 mg. g^{-1} F.W.) appeared in plants grown in full sun conditions compared to plants grown in shade conditions, which gave values of 1.94 and 2.00, respectively, in both seasons.

In the case of treatment with MeJA, it was found that it had a negative effect on the content of anthocyanin pigment in spikes, where the highest increase was found in untreated plants.

In the interaction treatments, it was found that plants not treated with MeJA gave the highest content of anthocyanin pigment, whether grown in conditions of full sunlight or in shade, but plants grown in the sun had a higher pigment content than plants grown in shade, and thus we conclude that MeJA had an negative effect on the formation of dye in flowers.

3.3.5. Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE) protein banding pattern

The polypeptides profiles upon of MeJA application with sunlight and shade revealed by SDS-PAGE are illustrated in Fig. (2) and Table (6). Demonstrative analysis of the presence and absence of polypeptides (bands) were assessed with (1) and (0), respectively, the total number of polypeptides was 9 with molecular weights ranging from 11.96 to 48.62 kDa. The highest number of bands was 9, detected in full sunlight + MeJA at 0, 0.05 and 0.1mM, the same number of bands appeared in the treatment shade + MeJA at 0 and 0.025 mM. While the lowest number of polypeptides appeared when exogenous application of MeJA at 0.1mM in shade treatment. SDS-PAGE results showed differences in patterns of protein between full sunlight and shade treatments either alone or in combination with exogenous MeJA different concentrations.



Fig. 2: SDS-PAGE protein banding patterns of *C. argentea* leaves (the arrows point to the disappearing bands)

Table 6: Data matrix illustrating the presence or absence of bands in the leaves of C. argentea

 protein electrophoresis banding patterns

	Full sunlight + MeJA (mM)					Shade+ MeJA (mM)				
MW	0.0	0.025	0.05	0.1	0.0	0.025	0.05	0.1		
48.62	1	1	1	1	1	1	1	1		
36.89	1	1	1	1	1	1	1	0		
31.39	1	1	1	1	1	1	1	1		
29.97	1	1	1	1	1	1	1	1		
26.72	1	1	1	1	1	1	1	1		
24.65	1	1	1	1	1	1	1	1		
21.84	1	0	1	1	1	1	0	1		
18.13	1	0	1	1	1	1	1	0		
11.96	1	1	1	1	1	1	1	0		
Total	9	7	9	9	9	9	8	6		

Our results revealed two bands uniquely (disappearance) expressed in response to application shade+ MeJA at 0.1mM with molecular weight 11.96 and 36.89 kDa while appeared in all others treatments. Polypeptide with molecular weight 21.84 kDa disappeared in treatments full sunlight + MeJA at 0.025 mM and shade+ MeJA at 0.05mM, as well as polypeptide 18.13 kDa disappeared in full sunlight + MeJA at 0.025 mM and shade+ MeJA at 0.1mM.

4. Discussion

Lighting is one of the important factors affecting plant growth and development, and its deficiency greatly affects plant performance and morphological characteristics. When the shading level is increased, plants adjust through a series of growth responses, such as increasing plant height, leaf hyponasty, leaf area and specific leaf area (Larbi *et al.*, 2015; Wu *et al.*, 2017).

It was observed from the obtained results that plants grown in the shade showed an increase in height compared to those grown in full sunlight, and this result agreed with (Thakur *et al.*, 2019; Haque *et al.*, 2009). Tan and Qian (2003) attributed this result to that the growth of plants in the shade leads to an increase in the production of gibberellic acid, which in turn leads to an increase in elongation of the cells without increasing their thickness, which gives the plant an etiolated appearance. In the event that the plant is exposed to shade, the plant tries to adapt to the conditions of lack of light by reducing the area of the leaves, and thus the respiration process decreases to help compensate for the significant decrease in the photosynthesis process (Campbell and Miller, 2002). This result has been proven in many studies (Thakur *et al.*, 2019).

The values of the rest of the vegetative traits, represented by stem diameter, number of leaves, number of branches, root length, and fresh and dry weight of both shoot and root, decreased. In the case of plant growth in the shade, this decrease may be due to that exposing the plant to shade led to a decrease in the accumulation of non-structural carbohydrates, which include soluble sugars and starch present in the leaves, which in turn help in the formation of structural carbohydrates, and thus the content of structural carbohydrates decreases, which in turn affects the formation of stem diameter and its stability (Hussain *et al.*, 2019). This result is consistent with what was obtained through our study as well.

Several studies have also mentioned that carbohydrates play an important role in encouraging plants to flower (Menzel and Simpson, 1988; Chen and Huang, 2005) and through our study, it was observed that the plant content of carbohydrates decreased in parallel with the decrease in the number of inflorescences produced in plants exposed to shade, and this was also proven by (Wu *et al.*, 2013; Yang *et al.*, 2014; Jian *et al.*, 2019) through their previous studies. Nielsen and Simonsen (2011) said that high light intensity promoted anthocyanin synthesis and accumulation. The anthocyanin contents of shaded plant leaves also significantly declined. Guo *et al.*, 2003; Yang *et al.*, 2005 reported that flower color of *P. lactiflora* faded as well, and anthocyanin concentration decreased dramatically in shaded plants; this was mostly due to a lack of soluble sugar supply, which was required to be translated into anthocyanin.

Under limiting light, plants set into motion a series of compensatory mechanisms, such as a substantial increase in photosynthetic pigments. This response allows the plant to maintain a photosynthetic antennae sufficient to capture the required light energy (Czeczuga, 1987) considering that highly pigmented leaves show a higher light absorption efficiency per leaf, which may allow the plant to achieve a carbon balance under light-deficit conditions (Dai *et al.*, 2009).

Flavonoids are one of the main components of phenolic compounds that mediate the antioxidant activity of phenolic compounds (Montero *et al.*, 2020). The highest content of total phenols, flavonoids and antioxidant activity was found in covered basil grown in the shade (Milenkovi'c *et al.*, 2021), this result confirms what was obtained through this study as the *C. argentea* plant content of flavonoids increased in shade-grown plants compared to plants growing in sunlight.

MDA, which is produced during lipid peroxidation, is an important index of cell damage under stress. Zhu *et al.* (2017) interpreted that the MDA content in purple pak-choi increased under low light stress, indicating that the degree of lipid peroxidation in the cell membrane is related to the duration of low light treatment and the degree of shaded light, and this is in line with the m obtained in our study as well.

Plants are adapted to survive under stress conditions by arresting their development and prioritizing defense mechanisms. This causes physiological and developmental changes in the plant. In all of these processes, hormones play a critical role (Guo *et al.*, 2018; Huot *et al.*, 2014). It has been demonstrated that the biosynthesis and accumulation of phytohormones such as abscisic acid (Jin and Chen, 2000), salicylic acid (Hoyos and Zhang, 2000), and jasmonate (Chao *et al.*, 1999; Thaler, 1999) form a pivotal adaptive strategy of plants to abiotic stresses. Hence, the application of phytohormones has been considered an effective way to improve plant growth and mitigate the effects of abiotic stress (Yan *et al.*, 2015).

Jasmonic acid (JA) and growth-related hormones antagonistically interact to coordinate plant growth and defense (Guo *et al.*, 2018). Both methyl jasmonate (MeJA) and jasmonic acid (JA), collectively referred to as jasmonates, are considered to have a beneficial effect on various crops subjected to environmental stress as mentioned by Yoon *et al.* (2009). For instance, exogenous MeJA has been reported to ameliorate the adverse effects of abiotic stress on chlorophyll concentration, photosynthetic rate, transpiration rate, proline content, and overall growth of plants (Yoon *et al.*, 2009).

Additionally, the application of exogenous JA can enhance stress tolerance by improving the antioxidant system, which includes enzymes and metabolites to scavenge the excessive ROSs generated as a result of various abiotic shocks (Avalbaev, 2016; Qiu *et al.*, 2014). exogenous application of MeJA increases the accumulation of osmolytes, such as proline, ABA, and soluble sugars, thus facilitating plants' adaptation to various abiotic stresses (de Ollas, 2013; Liu *et al.*, 2012; Ouli-Jun *et al.*, 2017; Sheteiwy *et al.*, 2018; Nowaz *et al.*, 2017; Sheteiwy *et al.*, 2021).

In earlier studies, it was shown that MeJA elicitor adversely affects cell growth in plant tissues and related in vitro cultures (Santamaria *et al.*, 2011). This might be explained by a MeJA-mediated decrease in the content of physiologically active cytokinins that are well known as senescence-inhibiting hormones. Further, MeJA has an inhibitory role on chlorophyll content and chloroplast transcriptional activity (Ananieva *et al.*, 2007).

The stronger overall stimulatory effect of Jasmonates on photosynthetic pigment accumulation could be due to its stronger effect on the chlorophyll synthesis pathway specially δ -ALA (aminolevulinic acid) which is the rate limiting step in the biosynthesis of chlorophyll during earliest stages of greening (Beale, 1978). Jasmonates treatment resulted in an increase of active cytokinin concentration which enhances chlorophyll accumulation in potato plant (Kovac and Ravnikar, 1994). Cytokinin is the phytohormones promoting light harvesting potential capacity of the plant, thus exerting a co-operative effect on the process of greening (Lew and Tsuji, 1982; Reiss C. and S. I. Beale, 1995).

Our results suggest that the inhibitory effect of MeJA on photosynthesis is actually due to the reduction of light-harvesting complexes, resulting in a reduction of the carbon fixation process. Down regulation of chlorophyll-related genes and chlorophyll-protein complexes has also been reported in previous studies, using genomic and proteomic tools to identify molecular changes in response to JA treatment (Agrawal, *et al.*, 2013, Chen *et al.*, 2011, Zhai *et al.*, 2007). In the presence of JA, synthesis of chloroplast proteins involved in photosynthesis was immediately reduced by negatively controlling translation, while transcript levels remained unchanged. The disappearance of the protein bands was attributed to the presence of the inhibitor with cycloheximide (an inhibitor of protein synthesis) and cordycepin (an inhibitor of mRNA synthesis) our results agreement with Ananieva and Ananiev (1999); Schenk *et al.* (2000); Sasaki *et al.* (2001) and Jung *et al.* (2007).

5. Conclusion

The light is an important and main factor to control in plant growth in health and greenish, it was clear from our results that *C. argentea* plants grown in the shade was affective negatively compared with the plants grown in the sunny area, when the plants forced to grow under shade condition we found that to avoid the harm effect of the shade by treating the plants with MeJA at concentration of 0.025 or 0.05mM.

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