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RESEARCH ARTICLE

Time course variations in ameliorative potentials of spermidine and kinetin for chromium toxicity - growth analysis indices of four *Vigna mungo* L. genotypes

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ABSTRACT

The experimental studies were performed for finding antagonistic role of PGRs viz., Kinetin a Polyamine (Spermidine) for soil supplied Chromium toxicity on mash genotypes with age. Four varieties i.e., 80, 88, 97 and ES-1 were sown. Earthen pots were used and placed in complete randomization arrangement. Chromium (Cr) doses were applied @ 30mg/kg and 60mg per kg soil. This was done by adding $CrCl_3$ salt in solutions form after 30 days of sowing. Spermidine and Kinetin were sprayed as 1.0 mM and 100.0 mM solutions respectively after 30 and 40 days of sowing. Growth analysis studies were carried when plants were of 30 and 46 days age. Chromium (Cr) at low level of concentrations in soil lowered the relative increase in plant height more effectively during growth interval 2 while, at higher toxicity levels, metal affected the shoot growth in the growth interval 1. Kinetin spray decreased the relative increase in plant height during growth interval 1while, Spermidine affected so during growth interval 1, increased the parameter. Chromium, at both levels of its concentration in soil, decreased the root growth rate more effectively during growth interval 2. Kinetin reduced relative increase in root length during growth interval 1 and increased it in the next growth interval. However, Spermidine effects started in the growth interval 1 and reached its maximum during growth interval 2. Chromium at lower concentration (30mg/kg soil) decreased the relative increase in leaf area during both intervals of growth. Exogenous Kinetin positively affected relative increase in leaf area and its effect being more pronounced in Ist interval of growth. Spermidine affected this attribute in the same manner but to lesser extent. Metal toxicity became effective during growth interval 2. Kinetin and Spermidine application to plants increased the net assimilation rate during both intervals comparatively more in growth interval 1.

Keywords: Chromium; Growth analysis; Kinetin; Leaf area; Root length; Spermidine; *Vigna*

INTRODUCTION

Heavy metals are contaminating agriculture soils and are emitted from various sources (Parveen et al., 2020; Rehman et al., 2020; Javed et al., 2020; Khalid et al., 2021). Microplastics are one of the sources of heavy metal for water pollution and affect the life of aquatic organisms (Khalid et al., 2020; Shah et al., 2021). When present in soils, heavy metals not only affect adversely the health of plant but also influence the soil microbes (Khalid et al., 2019; Shahid et al., 2019) in addition to human health through food chain (Zaheer et al., 2020). Heavy metals accumulations in soils or plants disturb the uptake and function of other mineral nutrients. Heavy metals toxicity generates Reactive oxygen species (ROS) which can intrupt many metabolic processes and growth of plants (Aqeel et al., 2021). Many remedial measures like metal removal, chelation or phytoextraction have been practiced to improve contaminated soils (Zaheer et al., 2020a). After it has been absorbed by the plant, adverse effects of heavy metal can be minimized through input of organic and inorganic compounds (Zaheer et al., 2020). Plant-microbe interaction is also an efficient method of Chromium detoxification due to its low cost (Sharma, et al., 2021). Chromium is a heavy metal which acts as trace element when present at its lower level of concentration but at high level, it acts as contaminant of the soil. Chromium is released by anthropogenic as well as natural resources into soil, air, and water leading to global environmental pollution (WHO, 2020).Chromium occurs as Cr (III) and Cr (IV) in

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environment. Both forms are absorbed by plants (Shahid, et al., 2017). Cr (VI) is absorbed actively through sulfate carriers (Xu, et al., 2021) while Cr (III) enters passively into plant cell (Singh, et al., 2013).

Kinetins regulate growth and developmental processes in plants (Sosnowski, 2019). Cell division, development, growth, nutrients uptake, assimilate and delaying of senescence are the phenomenon in which Kinetins are involved. Kinetins bind to membrane receptors starting a signal for transduction (Vyroubalova, et al., 2009). Kinetins enhance pigments concentrations and promote photosynthetic rate leading to more growth (Ahanger et al., 2019). Kinetins help in minerals nutrients uptake (khan et al., 2016). Polyamines perform wide array of roles in plant growth, development and stress tolerance (Chen et al., 2019). Exogenous polyamines application protects plant against environmental stresses (Chen et al., 2019) like salt (Jiang et al., 2020), metal (Yu et al., 2018) and water stress (Ebeed et al., 2017). Exposure of plant to stress increases its internal polyamines contents (Yu et al., 2019). Exogenous polyamine also protect photosynthetic machinery when enters into chloroplast (He et al., 2002).Among naturally occurring common polyamines, Spermidine is considered to be the most effective for stress tolerance (Shen et al., 2000). Reduces production of ROS and activates enzymatic and non-enzymatic antioxidant production are due to Spermidine (Nahar et al., 2017).

Growth analysis study of plant is an approach for measuring and evaluating the growth and productivity of plant (Wilson, 1981). Growth analysis studies help us to understand how plant can accumulate dry matter and what kinds of events make plant more productive (Ahad, 1986). Growth and leaf area of plants during development vary depending on plant species (Das Gupta & Nath, 2015; Kierzkowski et al., 2019). It also depends sometime on types of tissue layer (Fox et al., 2018). Environmental factors like water and nutrients supply, CO_2, temperature in addition to species, are the factors which regulate growth analysis parameters (Poorter et al., 2012; Tardieu et al., 1999; Shah et al., 2021). Growth rate of a plant is related to photosynthetic carbon assimilation (Fatichi et al., 2019) and sink strength (Wang et al., 2020). High SLA having species grows faster (Wright et al., 2004). For a plant, growth parameters like LAI (Leaf Area Index) and RGR (Relative Growth Rate) are important in determination of plant yield (Sun et al., 1999). Srivastava and Singh (1980) reported that growth process such as NAR (Net Assimilation Rate) and RGR (Relative Growth Rate) influence the yield of plant. Similarly, Thakur and Patel (1998) reported that dry matter production, LAI, NAR and RGR are responsible for high yield. Tesfaye et al., (2006) reported that when LAI is high, lowers evaporation decreases and maximum light is converted into dry matter. The growth and yield of a plant are regulated by plant metabolic activities and are affected by genetic and environmental factors.

Mash (*Vigna mungo* L.) is one of the most important pulse crops. Its seeds are used as whole or ground into powder which is used to make cake. Green seed and pods are cooked as vegetables. Fresh whole plant is used as manure. Dry plant is used for animal feed. Seeds contain considerable amount of protein, fats and carbohydrates as nutritive agents. Keeping in view the emerging sources of Chromium, its adverse impacts on plants, importance of Kinetin and polyamines in plant metabolism, this experimental study was managed for exploring the role of Kinetin and Spermidine to antagonize Chromium effects on mashbean genotypes. The objective was to explore the mitigating role of Kinetin and Spermidine for Chromium effects with the age of plant.

MATERIAL AND METHODS

This experiment was as pot culture to find out antagonistic effects both of exogenously sprayed Kinetin and a Ployamine (Spermidine) for toxicity of soil supplied Chromium (Cr) in pots. Four mash genotypes were grown in effluents free loamy soil. Mash genotypes 80, 88, 97 and ES-1 were obtained from a research institute (AARI) of Faisalabad (Pakistan). Salt of Chromium and PGRs used were of Sigma Company of Japan obtained from a dealer.

Chromium Chloride was supplied in pots when plants were of fifteen days. Pots without supplements of metal were considered control plants. PGRs (Spermidine @ 1.0mM and Kinetin @100.0mM) were sprayed twice using Tween-20 (0.1%) as surfactant after fifteen days repeates.

Four replicates from each genotype and treatments were evaluated three times with interval of 15 days starting from the day of PGRs spray completion.

Dry mass (DM) was determined after drying the plant parts in an oven at 65^{o}C to get constant weight. Following growth analysis parameters were evaluated according to method described by given against each.

Relative increase in plant height $=\ln h_2 - \ln h_1/T_2 - T_1$ (Radford, 1967)

Relative increase in root length $=\ln r_2 - \ln r_1/T_2 - T_1$ (Radford, 1967)

Relative increase in leaf area $= \ln L_2 - \ln L_1/T_2 - T_1$ (Radford, 1967)

Leaf area ratio; LAR= $L_2 + L_{1/}w_1 + w_2$ (West et al, 1920)

Relative leaf growth rate; RLGR = $\ln L_2 - \ln L_1$ (West et al, 1920)

Relative growth rate; RGR = $1/w_1 \times w_2/T_2-T_1$ (Radford, 1967)

Net assimilation rate NAR = $M_2-M_1/L_2-L_1 \times \ln L_2-\ln L_1/T_2-T_1$ (Gregary, 1917)

Where

M_1 is the initial total (shoot + root) dry mass, M_2 is the final total dry mass, L_1 is the initial leaf area, L_2 is the final leaf area, and (T_2 - T_1) is number of days between the two samplings.

Leaf area was calculated by measuring length and width of leaf with measuring scale. Dry biomass was determined by digital balance after drying plants in oven at 60^0C till getting constant weight.

RESULTS

Relative increase in plant height (cm cm^{-1} day^{-1})

Chromium at concentrations of 30mg/kg in soil lowered the relative increase in plant height more effectively during growth interval 2 (46-60 days age) [Table 1; column 4] while at higher toxicity levels (60mg/kg soil), metal affected the shoot growth in the growth interval 1 (30-45 days age) followed by decrease in their effects in subsequent growth interval [Table 1; column 7].

Kinetin [Table 1; column 2] spray decreased the relative increase in plant height during growth interval 1 (30-45 days age). While, Spermidine [Table 1; column 3] spray during growth interval 1 (30-45 days age) increased the parameter followed by decrease in next growth interval (46-60 days age). Exogenous application of Kinetin and Spermidine added their effects to Chromium toxicity in decreasing plant growth rate [Table 1; column 5, 6]

Relative increase in root length (cm cm^{-1} day^{-1})

Chromium, at both levels of its pollution in soil, decreased the root growth rate more effectively during growth interval 2 (46-60 days age) except V_3 (MASH 97) and V_4 (MASH ES-1) where it affected the root growth during the growth interval 1 (30-45 days age) followed by an increase in the subsequent growth interval [Table 2; column 4 and 7]. Kinetin and Spermidine, in general, increased the rate of root growth. Kinetin slightly inhibited relative increase in root length during growth interval 1 (30-45 days age) followed by its increase in the next growth interval [Table 2; column 2]. However, Spermidine effects started in the growth interval 1 and reached its maximum during growth interval 2 (46-60 days age). Spermidine was found to be more effective than Kinetin in playing its positive role for relative increase in root length.

Exogenous application of Kinetin and Spermidine alleviated the toxic effects of metals on plants by influencing plant root growth at the same growth intervals as were effective for plants of non polluted soils [Table 2; column 5, 6].

Relative increase in leaf area (cm^2 cm^{-2} day^{-1})

Chromium at lower concentration (30mg/kg soil) decreased the relative increase in leaf area during two intervals of growth (30-60 days age) except in V_4 (MASH ES-1) in which this effect was limited only to growth interval 1 (30-45 days age) [Table 3; column 4]. Plants grown in higher Chromium concentration (60mg/kg soil) exhibited decrease in this attribute through entire growth period [Table 3; column 7].

This was comparatively to a greater extent than lower level of Chromium. Exogenous Kinetin positively

Table 1: Relative increase in plant height [$\ln h_2 - \ln h_1/t_2 - t_1$; (cm cm^{-1} day^{-1})] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. Sprays
V_1	$H_2 - H_1$	0.029	0.026	0.034	0.023	0.021	0.027	0.015	0.012	0.020
	$H_3 - H_2$	0.021	0.021	0.024	0.021	0.019	0.023	0.020	0.015	0.020
V_2	$H_2 - H_1$	0.032	0.029	0.040	0.027	0.022	0.030	0.020	0.019	0.021
	$H_3 - H_2$	0.025	0.027	0.026	0.018	0.017	0.021	0.022	0.015	0.025
V_3	$H_2 - H_1$	0.034	0.030	0.041	0.027	0.022	0.031	0.022	0.020	0.022
	$H_3 - H_2$	0.023	0.025	0.024	0.018	0.018	0.019	0.019	0.012	0.020
V_4	$H_2 - H_1$	0.032	0.027	0.039	0.024	0.018	0.027	0.018	0.017	0.018
	$H_3 - H_2$	0.025	0.028	0.027	0.018	0.018	0.019	0.012	0.009	0.007

H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)] $H_2 - H_1$=Growth interval 1; $H_3 - H_2$=Growth interval 2. h_2-h_1=plant height difference of harvests; t2-t1=time interval between harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine. Calculations are based on log-transformed means to achieve homogenous distribution of residuals.

Table 2: Relative increase in root length [$\ln r_2 - \ln r_1/t_2 - t_1$; ($cm\ cm^{-1}\ day^{-1}$)] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. Sprays	Spd. Sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. Sprays
V_1	$H_2 - H_1$	0.051	0.050	0.055	0.042	0.037	0.047	0.029	0.028	0.035
	$H_3 - H_2$	0.013	0.010	0.019	0.005	0.006	0.008	0.002	0.003	0.005
V_2	$H_2 - H_1$	0.047	0.041	0.048	0.035	0.029	0.039	0.022	0.021	0.026
	$H_3 - H_2$	0.007	0.008	0.015	0.004	0.007	0.004	0.002	0.004	0.005
V_3	$H_2 - H_1$	0.035	0.028	0.035	0.014	0.009	0.021	0.008	0.007	0.012
	$H_3 - H_2$	0.016	0.018	0.027	0.027	0.025	0.029	0.047	0.033	0.041
V_4	$H_2 - H_1$	0.038	0.029	0.037	0.021	0.015	0.024	0.015	0.009	0.014
	$H_3 - H_2$	0.012	0.014	0.024	0.017	0.020	0.024	0.025	0.014	0.031

H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)]. $H_2 - H_1$=Growth interval 1; $H_3 - H_2$=Growth interval 2. r_2-r_1=root length difference of harvests; t2-t1=time interval between harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine. Calculations are based on log-transformed means to achieve homogenous distribution of residuals.

Table 3: Relative increase in leaf area [$\ln L_2 - \ln L_1/t_2 - t_1$; ($cm^2\ cm^{-2}\ day^{-1}$)] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays
V_1	$H_2 - H_1$	0.0070	0.0224	0.0130	-0.0074	0.0043	-0.0074	-0.0280	-0.0218	-0.0313
	$H_3 - H_2$	0.0028	0.0068	0.0049	-0.0016	0.0080	0.0060	-0.0050	0.0038	0.0083
V_2	$H_2 - H_1$	0.0077	0.0104	0.0121	-0.0023	0.0079	0.0097	-0.0167	0.0005	-0.0187
	$H_3 - H_2$	0.0039	0.0078	0.0070	-0.0003	0.0079	0.0064	-0.0046	0.0019	0.0074
V_3	$H_2 - H_1$	0.0040	0.0203	0.0097	0.0004	0.0097	0.0002	-0.0244	-0.0012	-0.0171
	$H_3 - H_2$	0.0090	0.0081	0.0053	0.0017	0.0052	0.0033	0.0020	0.0010	0.0038
V_4	$H_2 - H_1$	0.0049	0.0106	0.0070	-0.0249	0.00104	-0.0125	-0.0103	-0.0039	-0.0367
	$H_3 - H_2$	0.0014	0.0088	0.0024	0.0054	0.0065	-0.0014	0.0043	0.0083	0.0109

H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)]. $H_2 - H_1$=Growth interval 1; $H_3 - H_2$=Growth interval 2. L_2-L_1=leaf area difference of harvests; t2-t1=time interval between harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine. Calculations are based on log-transformed means to achieve homogenous distribution of residuals.

affected relative increase in leaf area except negative effects in V_2 (MASH 88) and V_3 (MASH 97) during growth interval 2 (46-60 days age). Kinetin was more effective in Ist interval of growth (30-45 days age) [Table 3; column 2 and 7]. Spermidine affected this attribute in the same manner but to lesser extent [Table 3; column 3]. Kinetin lowered the toxic effects of Chromium [Table 3; column 5 and 8]. This was more obvious during later interval of growth. Such influence of Kinetin was dependent on Chromium concentration. Spermidine showed similar effect but to lower degree and restricted mainly to 2nd growth interval (46-60 days age) [Table 3; column 6and 9].

Leaf area ratio (LAR) ($cm^2\ g^{-1}$)

Chromium toxicity decreased the leaf area ratio in concentration dependent manners except V_4 (MASH ES-1) where higher level of Chromium toxicity slightly increased the leaf area ratio [Table 4; column 4 and 7].

Kinetin and Spermidine also decreased the leaf area ratio in polluted [Table 4; column 4] and non polluted [Table 4; column 2 and 3] soil grown plants. Spermidine has more decreasing effects than Kinetin on plants of non polluted soil and vice versa.

Relative leaf growth rate (RLGR) (cm^2)

Both Chromium levels stress decreased the relative leaf growth rate, effect being more pronounced during growth interval 2 (46-60 days age). Exogenous application of Kinetin alleviated the toxic effects of low Chromium level (30mg/kg soil) during growth interval 2 except V_1 (MASH 80) [Table 5; column 4].

Spermidine was effective in mitigating the toxicity of low Chromium level (30mg/kg soil) during the Ist growth intervals in MASH 80 and MASH 88 [Table 5; column 6].

Relative growth rate (RGR) ($g\ g^{-1}\ day^{-1}$)

Low Chromium concentration (30mg/kg soil) exerted negative effects on relative growth rate in the 2nd interval of growth (46-60 days age), except V_4 (MASH ES-1). Unlike low level of Chromium, higher level (60mg/kg soil) Chromium started its induction to destructiveness, with comparatively more severity, during growth interval 1(30-45 days age) except V_3 (MASH 97) [Table 6; column 7].

Plants grown in polluted soil reflected limitation in relative growth rate. Chromium at higher concentration was proved to be more toxic. Effects being more pronounced during Ist growth interval, exogenous application of Kinetin

Table 4: Leaf area ratio [LAR=L_1+L_2/W_1+W_2); (cm^2 g^{-1})] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. Sprays	Spd. sprays
V_1	H_2 - H_1	45.88	49.76	38.63	36.25	22.96	22.35	34.00	19.99	22.83
	H_3 - H_2	31.35	36.86	26.39	22.83	15.10	13.22	17.70	9.62	10.96
V_2	H_2 - H_1	37.98	35.49	30.91	31.06	19.53	21.65	30.21	19.77	20.64
	H_3 - H_2	26.08	23.70	21.08	20.76	13.17	15.06	17.75	11.88	11.23
V_3	H_2 - H_1	36.64	35.02	33.60	31.63	27.60	23.72	25.83	29.50	28.72
	H_3 - H_2	26.54	25.93	33.03	22.52	19.74	15.42	14.51	19.38	17.36
V_4	H_2 - H_1	32.34	30.71	32.95	27.33	24.87	22.32	42.95	26.79	26.26
	H_3 - H_2	23.33	22.83	23.62	18.00	17.96	13.38	49.12	19.94	15.77

H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)]. H_2 - H_1=Growth interval 1; H_3 - H_2=Growth interval 2. W_1+W_2=dry mass of harvests; L_1+L_2=leaf area of harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine.

Table 5: Relative leaf growth rate [RLGR=lnL_2 - lnL_1; (cm^2)] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays
V_1	H_2 - H_1	1.031	1.001	1.093	0.851	0.748	0.945	0.598	0.552	0.709
	H_3 - H_2	0.261	0.201	0.375	0.106	0.124	0.169	0.044	0.064	0.010
V_2	H_2 - H_1	0.954	0.823	0.965	0.707	0.575	0.773	0.451	0.415	0.519
	H_3 - H_2	0.139	0.163	0.311	0.085	0.145	0.074	0.035	0.079	0.109
V_3	H_2 - H_1	0.709	0.555	0.701	0.292	0.177	0.437	0.163	0.135	0.242
	H_3 - H_2	0.323	0.368	0.551	0.551	0.500	0.577	0.935	0.664	0.818
V_4	H_2 - H_1	0.761	0.586	0.739	0.416	0.295	0.485	0.295	0.198	0.284
	H_3 - H_2	0.239	0.272	0.487	0.345	0.398	0.485	0.501	0.288	0.620

V_1 (MASH 80); V_2 (MASH 88); V_3 (MASH 97); V_4 (MASH ES-1); ppm=mg/kg or µg/g; Kin=Kinetin; Spd=Spermidine. H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)]. H_2 - H_1=Growth interval 1; H_3 - H_2=Growth interval 2. L_2-L_1=leaf area differences of harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine. Calculations are based on log-transformed means to achieve homogenous distribution of residuals.

Table 6: Relative growth rate [RGR=1/w_1×w_2/t_2-t_1; (g g^{-1} day^{-1})] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays
V_1	H_2 - H_1	0.0206	0.0283	0.0391	0.0279	0.0677	0.0621	0.0194	0.0613	0.0477
	H_3 - H_2	0.0258	0.0286	0.0214	0.0120	0.0078	0.0069	0.0103	0.0088	0.0087
V_2	H_2 - H_1	0.0232	0.0309	0.0425	0.0309	0.0699	0.0646	0.0228	0.0643	0.0498
	H_3 - H_2	0.0253	0.0282	0.0210	0.0108	0.0078	0.0069	0.0098	0.0081	0.0096
V_3	H_2 - H_1	0.0208	0.0386	0.0325	0.0297	0.0468	0.0504	0.0283	0.0337	0.0245
	H_3 - H_2	0.0241	0.0229	0.0225	0.0102	0.0112	0.0081	0.0077	0.0128	0.0132
V_4	H_2 - H_1	0.0142	0.0238	0.0146	0.0050	0.0324	0.0308	-0.0616	0.0226	0.00096
	H_3 - H_2	0.0229	0.0248	0.0257	0.0133	0.0123`	0.0097	0.0312	0.0132	0.0168

V_1 (MASH 80); V_2 (MASH 88); V_3 (MASH 97); V_4 (MASH ES-1); ppm=mg/kg or µg/g; Kin=Kinetin; Spd=Spermidine. H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)]. H_2 - H_1=Growth interval 1; H_3 - H_2=Growth interval 2. W2-W1=dry mass difference of harvests; t_2-t_1=time interval between harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine. Calculations are based on log-transformed means to achieve homogenous distribution of residuals.

improved the relative growth rate of plants through all growth intervals, except V_1 (MASH 80) [Table 6; column 2]. Spermidine application had a promotive effect on relative growth rate with a greater degree of extent than that of Kinetin but limited only to the Ist interval of growth. In the next growth intervals Spermidine could not had marked influence or response was either negligibly inverse to this [Table 6; column 3]. Kinetin and Spermidine added their effectiveness to Chromium effects for increasing relative growth rate but this was limited only to Ist growth interval (30-45 days age). In the subsequent growth phases, both PGRs showed enhancement effect to Chromium toxicity for decrease in this attribute. Such behavior of PGRs was dominant at lower Chromium toxicity (30mg/kg soil) [Table 6; column 5, 6, 8 and 9].

Net assimilation rate (NAR) (mg cm^{-2})

Metal decreased the net assimilation rate at all applied concentration. However, metals did not affect the net assimilation rate during Ist growth interval (30-45 days age). Metal toxicity became effective during growth interval 2 (37-52 days age). Kinetin application to plants increased the net assimilation rate starting its effect in growth interval 1 and not vanishing but fading in the subsequent interval of growth [Table 7; column 2]. A similar and much more effective influence was shown by Spermidine for this variable [Table 7; column 3].Plants showed increase in net assimilation rate when subjected to Kinetin and low Chromium concentration. Increase was much more than caused by mere application of Chromium in early growth stage (30-45 days age). This revealed the role of Kinetin in adding the positive effects to Chromium for net assimilation rate. Slight or no increase in this attribute due to exogenous Kinetin supply was noted for the remaining interval of plant growth [Table 7; column 5].At higher Chromium concentration (60mg/kg soil), Kinetin application improved the values for net assimilation rate [Table 7; column 8].

Effects of Spermidine spray in increasing net assimilation rate were similar by interval to that manifested by Kinetin but to a lesser extent [Table 6; column 6].The effects of Kinetin and Spermidine in increasing net assimilation rates of plants grown in toxic environment were in accordance with the severity of metal stress.

DISCUSSION

Growth analysis studies have a long history of evaluating plant growth and physiology. According to Hall and Long (1993), it is much better to study plant as a whole rather than study of its isolated parts. Initially, the growth indices given by West et al. in 1920 were aimed to resolve the plant growth phenomenon in term of dry mass production. Gregory (1917) was the first to design growth indices which were later on referred as unit leaf growth rate. Leaf growth expresses growth as increase in foliar surface and is a key factor for evaluation of plant performance (Hilty et al., 2021). Environmental factors like water and nutrients supply, CO_2, temperature in addition to species are the factors which regulate growth analysis parameters (Poorter et al., 2012; Tardieu et al., 1999; Shah et al., 2021).

In our experiment, the results revealed that Chromium at low level lowered relative increase in plant height, root length and leaf area more effectively during interval 2 while higher level was effective in the growth interval 1. Kinetin spray decreased the relative increase in plant height during growth interval 1. Spermidine spray increased the parameters during growth interval 1. Chromium effects on shoot length and plant biomass have been reported by many researchers (Ding, et al., 2019; Shiyab, 2019). Kinetin mediates increase in plant height (Wu Zhen Ling et al., 1998). Foliar spray of polyamine increased the height. Chromium effects on plant root as causing root growth retardation are reported (Kakkalameli, et al., 2018). It has been reported that Kinetin treatment results in alleviation of the negative effects of heavy metals on the root growth and stem growth (Khafaga et al., 1997). A positive correlation between polyamine and plant root growth and induction of adventitious root formation has been reported also (Neves et al., 2002). Chromium toxicity reduces leaf area and leaf number (Vernay et al., 2007). Application of Kinetin resulted in alleviation of negative effects of cadmium on plant leaf area (Haroun, et al., 2003).

In our experiment, Chromium decreased the net assimilation rate significantly during growth interval 2. Kinetin application to plants increased the net assimilation rate starting its effect in growth interval 1. In the present results, exceptions and variation in the responses of varieties might be due to their genetic differences. Net assimilation rate is the capacity of plant form dry weight as a leaf area. This term explains photosynthetic potential and in combination with the leaf area ratio (LAR) and relative growth rate (RGR)

Table 7: Net assimilation rate [NAR=M2-M1/L_2-L_1×lnL_2-lnL_1/t_2-t_1; (mg cm^{-2})] of mash exposed to metal toxicity, kinetin and Spermidine

	Diff. between harvests	No metal input			Cr (30ppm)			Cr (60ppm)		
		Dist. H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. sprays	Spd. sprays	Dist.H_2O sprays	Kin. Sprays	Spd. Sprays
V_1	H_2 - H_1	0.00044	0.00056	0.00097	0.00075	0.00256	0.00247	0.00057	0.00277	0.00201
	H_3 - H_2	0.00080	0.00075	0.00080	0.00052	0.00052	0.00052	0.00058	0.00092	0.00080
V_2	H_2 - H_1	0.00060	0.00084	0.00130	0.00096	0.0031	0.0026	0.00075	0.0028	0.0022
	H_3 - H_2	0.00095	0.00116	0.00098	0.00051	0.00059	0.00045	0.00055	0.00068	0.00086
V_3	H_2 - H_1	0.00056	0.00106	0.00093	0.00091	0.00158	0.00196	0.00109	0.00110	0.00084
	H_3 - H_2	0.00089	0.00087	0.00096	0.00045	0.00057	0.00052	0.00053	0.00066	0.00075
V_4	H_2 - H_1	0.00043	0.00076	0.00044	0.00018	0.00126	0.00134	0.00131	0.00083	0.00071
	H_3 - H_2	0.00096	0.00107	0.00106	0.00074	0.00068	0.00072	0.00061	0.00066	0.00106

V_1 (MASH 80); V_2 (MASH 88); V_3 (MASH 97); V_4 (MASH ES-1); ppm=mg/kg or μg/g; Kin=Kinetin; Spd=Spermidine. H_1=Ist harvest [on the day of PGRs application (30 days age)]; H_2=2nd harvest [15 days after H_1 (46 days age)]. H_3=3rd harvest [15 days after H_2 (60 days age)]. H_2 - H_1=Growth interval 1; H_3 - H_2=Growth interval 2. M_2-M_1=dry mass difference of harvests; L_2-L_1=leaf area differences of harvests; t_2-t_1=time interval between harvests. V_1 (80); V_2 88); V_3 (97); V_4 (ES-1); ppm=mg per kg ; Kin=Kinetin; Spd=Spermidine. Calculations are based on log-transformed means to achieve homogenous distribution of residuals.

it is used for analysis of plant response to environmental factors. Net assimilation rate depends upon growth of leaf and dry mass production. These characters are affected by photosynthesis, respiration and water potential of plant etc. Water potential affects stomatal opening and cell growth finally changing leaf expansion rate. A reduction in leaf growth ultimately affects net assimilation rate (NAR), LAR and RGR. Relative growth rate is the product of leaf area ratio and net assimilation rate RGR = LAR× NAR. Relative growth rate and net assimilation rate of plants are changed by various factors treatments.

The inhibitory effects of heavy metals are more on roots because of direct contact and higher metal accumulation in them (Breckle, 1991). Inhibition of plant RGR treated with metal mainly might be due to decreased NAR. Previously, changes in LAR, LMR, and SLA were also observed by metal treatment (Abo-Kassem et al., 1995). The negative effect of metal on plant is also in the form of an increase in dry to fresh weight ratio (DM/FM) in all plant organs (Moya et al., 1993).

Relative growth rate is dependent on NAR and leaf area ratio (Lambers et al., 1989: Shiply, 2006) which are the physiological and morphological components of RGR respectively. In turn, LAR depends on specific leaf area (SLA) and leaf mass ratio (LMR). NAR is not expression of real photosynthesis because it tells us about the net gain of photosynthesis after consumptions in respiration. As leaf area is likely to increase with plant age but LAR decreases due to leaf fall in older plant so that NAR can be irrespective photosynthesis change.

Temporal variations in growth pattern of plant are conducive to change in its metabolic activity which in turn depends upon, mainly, the activity of enzymes. A connection between physiological activities and dark reaction exists in plant. Dark respiration regulates cell metabolism. Half of the photosynthates synthesized every day are consumed in respiration in the same period. Hence, dark respiration has a key importance in biomass production (Lambers, 1985).

The effects of heavy metals on dark respiration either may be early or relatively late. This difference is due to metal uptake of plant until a critical metal concentration is reached. The early effect is to accelerate dark respiration. During this stage the control of cell on metabolism remains stable (Van Assche et al., 1988) under stress. Lee et al., (1976) reported increase in dark respiration and activity of enzymes like glutamate dehydrogenase, isocitrate dehydrogenase and malate dehydrogenase. According to Van Assche et al., (1988) the activity of malate enzyme and enzymes of oxidative pentose phosphate pathway, glucose-6-phosphate dehydrogenase, is increased under metal treatment to compensate NADPH deficiency. Ernst (1980) reported that enhanced dark respiration is a compensation mechanism for supply of ATP by oxidative phosphorylation. Production of ROS and antioxidant enzymes during stress also affects dark respiration (Okuda et al., 1991). The late effect of metal is expressed on dark respiration inhibition and its enzymes (Oliveira et al., 1994). According to Van Assche et al., (1988), physiological state of cell is reversed when heavy metal concentrations exceed its toxic limit. Metals inhibit activity of enzymes like ATPase and others through binding SH domain. It may change cation balance at the subcellular levels also (Lindberg and Wingstrand 1985; Fodor et al., 1996). By this action of metals, changes in photosynthetic rate occur which contribute in growth changes.

Leaf area ratio (LAR) reduction can be as consequence of low leaf mass ratio (LMR) and specific leaf area (SLA). Under metal stress no significant change in LMR occurs. Barcelo et al., (1988) reported that the key reason for LAR reduction could be low SLA by a disorder in water supply ratio. The reduction in water uptake can be due to root growth inhibition (Marchiol et al., 1996).

CONCLUSION

The effect of Chromium was more during early growth stage of plant while Kinetin and Spermidine were found to be more influential during late growth stages.

Conflict of interest

Author has no conflict of interest and there was no funding body for research and manuscript.

Authors contribution

Ghulam Yasin designed and conducted experiment. Saira Sameen analysed the data. Shahzadi Saima, Ikram ul Haq and Adeela Altaf participated in manuscript drafting and proof reading.

REFERENCES

Abo-Kassem, E., A. Sharaf-El-Din, J. Rozema and E. Foda. 1995. Synergistic effects of cadmium and NaCl on the growth, photosynthesis and ion content in wheat plants. Biol. Plant. 37: 241-324.

Ahad, M. A. 1986. Growth Analysis of Rice Bean (*Vigna umbellata* Thunb.) Under Different Management Practices and their Agronomic Appraisal. Ph.D. Dissertation in Agronomy University of Nebraska, Lincoln, United States, p21-22.

Ahanger, M. A., U. Aziz, A. A. Alsahli, M. N. Alyemeni and P. Ahmad. 2019. Combined kinetin and spermidine treatments ameliorate growth and photosynthetic inhibition in *Vigna angularis* by up-regulating antioxidant and nitrogen met abolism under cadmium stress. Biomology. 10: 147.

Aqeel, M., N. Khalid, A. Tufail, R. Z. Ahmad, M. S. Akhter, M. Luqman, M. T. Javed, M. K. Irshad, S. Alamri, M. Hashem and A. Noman A. 2021. Elucidating the distinct interactive impact of cadmium and nickel on growth, photosynthesis, metal homeostasis, and yield responses of mung bean (*Vigna radiata* L.) varieties. Environ. Sci. Pollut. Res. Int. 28: 27376-27390.

Barcelo, J., M. D. Vazquez and C. Poschenrieder. 1988. Cadmium induced structural and ultrastructural changes in the vascular system of bush bean stems. Acta Bot. 101: 254-261.

Breckle, S. 1991. Growth under stress. Heavy metals. In: (Eds.), Plant Poots, The Hidden Half. Dekker, Inc., New York, p351-373.

Chen, D., Q. Shao, L. Yin, A. Younis and B. Zheng. 2019. Polyamine function in plants, met abolism, regulation on development, and roles in abiotic stress responses. Front. Plant Sci. 9: 1945.

Das Gupta, M. and U. Nath. 2015. Divergence in patterns of leaf growth polarity is associated with the expression divergence of miR396. Plant Cell. 27: 2785-2799.

Ding, G., Z. Jin, Y. Han, P. Sun, G. Li and W. Li. 2019. Mitigation of chromium toxicity in *Arabidopsis thaliana* by sulfur supplementation. Ecotox. Environ. Saf. 182: 109379.

Ebeed, H. T., N. M. Hassan and A. M. Aljarani. 2017. Exogenous applications of polyamines modulate drought responses in wheat through osmolytes accumulation, increasing free polyamine levels and regulation of polyamine biosynthetic genes. Plant Physiol. Biochem. 118: 438-448.

Ernst, W. 1980. Biochemical aspects of cadmium in plants. In: Cadmium in the Environment. Wiley and Sons, New York, p639-653.

Fatichi, S., C. Pappas, J. Zscheischler and S. Leuzinger. 2019. Modelling carbon sources and sinks in terrestrial vegetation. New Phytol. 221: 652-668.

Fodor, F., E. Sarvari, F. Lang, Z. Szigeti and E. Cseh. 1996. Effects of Pb and Cd on cucumber depending on the Fe-complex in the culture solution. J. Plant Physiol. 148: 434-439.

Fox, S., P. Southam, F. Pantin, R. Kennaway, S. Robinson, G. Castorina, Y. E. Sanchez-Corrales, R. Sablowski, J. Chan and V. Grieneisen. 2018. Spatiotemporal coordination of cell division and growth during organ morphogenesis. PLoS Biol. 16: e2005952.

Gregory, F. G. 1917. Physiological Conditions in Cucumber Houses. Experimental Research Stationn Cheshunt 3rd Annual Report, United Kingdom, p19.

Hall, D. and S. Long. 1993. Photosynthesis and the changing environment. In: Photosynthesis and Production in a Changing Environment. In: D. Hall, H. Bolhar-Nordenkampf, R. Leegood and S. Long, (Eds.), A Field and Laboratory Manual Introduction. Chapman & Hall, United Kingdom.

Haroun, S. A., H. S. Aldesuquy, S. A. Abo-Hamed and O. P. El-Said. 2003. Kinetin-induced modification in growth criteria, ion contents and water relations of sorghum plants treated with cadmium chloride. Acta Bot. Hung. 45: 113-126.

He, L. X., K. Nada, Y. Kasukabe and S. Tachibana. 2002. Enhanced susceptibility of photosynthesis to low-temperature photoinhibition due to interruption of chill- induced increase of S-adenosylmet hionine decarboxylase activity in leaves of spinach (*Spinacia oleracea* L.). Plant Cell Physiol. 43: 196-206.

Hilty, J., B. Muller, F. Pantin and S. Leuzinger. 2021. Plant growth: The what, the how, and the why. New Phytol. 232: 25-41.

Javed, M. T., H. H. Saleem, S. Aslam, M. Rehman, N. Iqbal, R. Begum, S. Ali, A. A. Alsahli, M. N. Alyemeni and L. Wijaya. 2020. Elucidating silicon-mediated distinct morpho-physio-biochemical attributes and organic acid exudation patterns of cadmium stressed Ajwain (*Trachyspermum ammi* L.). Plant Physiol. Biochem. 157: 23-37.

Jiang, D. X., X. Chu, M. Li, J. I. Hou, X. Tong, Z. P. Gao and G. X. Chen. 2020. Exogenous spermidine enhances salt-stressed rice photosynthetic performance by stabilizing structure and function of chloroplast and thylakoid membranes. Photosynth. 58: 61-71.

Kakkalameli, S. B., A. Daphedar, N. Hulakoti B. N. Patil and T. C. Taranath. 2018. *Azolla filiculoides* lam as a phyto tool for remediation of heavy metals from sewage. Int. J. Pharm. 8: 282-287.

Khafaga, E. R., A. M. Abed and R. A. Agamy. 1997. Effect of kinetin and decapitation on growth and yield of Egyptian clover (*Trifolium alexandrinum* L. I.) Effect on morphological and anatomical characters. Bull. Faculty Agric. Univ. Cairo. 48: 283-308.

Khalid, H., M. Zia-ur-Rehman, A. Naeem, M. U. Khalid, M. Rizwan, S. Ali, M. Umair, and M. I. Sohail. 2019. *Solanum nigrum* L. Novel hyperaccumulator for the phytomanagement to cadmium contaminated soils. In: Cadmium Toxicity and Tolerance in Plants; Elsevier, Amsterdam, Netherlands, p451-477.

Khalid, N., Z. F. Rizvi, N. Yousaf, S. M. Khan, A. Noman, M. Aqeel, K. Latif and A. Rafique. 2021. Rising metals concentration in the environment: A response to effuents of leather industries in Sialkot. Bull. Environ. Contam. Toxicol. 106: 493-500.

Khalid, N., M. Aqeel, A. Noman, M. Hashem, Y. Mostafa, H. A. S. Alhaithloul and S. M. Alghanem. 2020. Linking effects of microplastics to ecological impacts in marine environments. Chemosphere. 264: 128541.

Kierzkowski, D., A. Runions, F. Vuolo, S. Strauss, R. Lymbouridou, A. L. Routier-Kierzkowska, D. Wilson-Sanchez, H. Jenke, C. Galinha, G. Mosca, Z. Zhang, R. D. Ioio, P. Huijser, R. S. Smith and M. Tsiantis. 2019. A growth-based framework for leaf shape development and diversity. Cell. 177: 1405-1418.

Lambers, H. 1985. Respiration in intact plants and tissues, its regulation and dependance on environmental factors, metabolism and invaded organism. In: , (Eds.), Encyclopedia of Plant Physiology, New Series. Springer-Verlag, Berlin, p418-473.

Lambers, H., N. Freijsen, H. Poorter, T. Hirose and A. Van der Werf. 1989. Analyses of growth based on net assimilation rate and nitrogen productivity; their physiological background. In: , (Ed.), Causes and Consequences of Variation in Growth Rate and Productivity of Higher Plants. Academic Publishing BV, Hague, Netherlands, p1-17.

Lee, S. M., D. H. Han, C. H. Lee and S. B. Kim. 1996. Effects of ABA and kinetin treatment on the coloration and quality of Campbell early and black olympiagrapes. J. Korean Soc. Hort. Sci. 37: 263-268.

Lindberg, S. and G. Wingstrand. 1985. Mechanism of Cd^{+2} inhibition of (K^+ + Mg^{2+}) ATPase activity and K^+ ($^{86}Rb^+$) uptake in young roots of sugar beet (*Beta vulgaris*). Physiol. Plant. 63: 181-186.

Marchiol, L., L. Leita, M. Martin, A. Peterssotti, G. Zerbi. 1996. Physiological responses of two soybean cultivars to cadmium. J. Environ. Qual. 25: 562-566.

Moya, J., R. Ros, I. Picazo. 1993. Influence of cadmium and nickel on growth, net photosynthesis and carbohydrate distribution in rice plants. Photosynth. Res. 36: 75-80.

Nahar, K., M. Hasanuzzaman, T. Suzuki and M. Fujit. 2017. Polyamines-induced aluminum tolerance in mung bean, a study on antioxidant defense and methylglyoxal detoxification systems. Ecotoxicology. 26: 58-73.

Neves, C., H. Santos, L. Vilas-Boas and S. Amâncio. 2002. Involvement of free and conjugated polyamines and free amino acids in the adventitious rooting of micropropagated cork oak and grapevine shoots. Plant Physiol. Biochem. 40: 1071-108.

Okuda, T., Y. Matsuda, A. Yamanaka and S. Sagisaka. 1991. Abrupt increase in the level of hydrogen peroxide in leaves of winter wheat in caused by cold treatment. Plant Physiol. 97: 1265-1267.

Oliveira, J., M. Oliva, J. Cambraia and Vanegas. 1994. Absorption, accumulation and distribution of cadmium by two soybean CVS. Rev. Brasil. Fisiol. Veget. 6: 91-95.

Parveen, A., M. H. Saleem, M. Kamran, M. Z. Haider, J. T. Chen, Z. Malik, M. S. Rana, A. Hassan, G. Hur and M. T. Javed. 2020. Effect of citric acid on growth, ecophysiology, chloroplast ultrastructure and phytoremediation potential of jute (*Corchorus capsularis* L.) Seedlings Exposed to Copper Stress. Biomol. 10: 592.

Poorter, H., K. J. Niklas, P. B. Reich, J. Oleksyn, P. Poot and L. Mommer. 2012. Biomass allocation to leaves, stems and roots: Meta-analyses of interspecific variation and environmental control. New Phyt. 193: 30-50.

Radford, P. J. 1967. Growth analysis formulae, their uses and abuses. Crop Sci. 7: 171-175.

Rehman, M., M. H. Saleem, S. Fahad, Z. Maqbool, D. Peng, G. Deng and L. Liu. 2020. Medium nitrogen optimized *Boehmeria nivea* L. growth in copper contaminated soil. Chemosph. 266: 128972.

Shah, F., S. Shah, A. Muhammad, S. Osman, W. Depeng, W. Chao and T. Veysel. 2021. Climate Change and Plants: Biodiversity, Growth and Interactions. Routledge, United Kingdom.

Shahid, M., M. T. Javed, A. Mushtaq, M. S. Akram, F. Mahmood, T. Ahmed, M. Noman and M. Azeem. 2019. Microbe-mediated mitigation of cadmium toxicity in plants. In: Cadmium Toxicity and Tolerance in Plants; Elsevier, Amsterdam, Netherlands. p427-449.

Shahid, M., S. Shamshad, M. Rafiq, S. Khalid, I. Bibi, N. K. Niazi, C. Dumat and M. I. Rashid. 2017. Chromium speciation, bioavailability, uptake, toxicity and detoxification in soil-plant system, A review. Chemosph. 178: 513-533.

Sharma, P., S. Tripathi, P. Chaturvedi, D. Chaurasia and R. Chandra. 2021. Newly isolated Bacillus sp. PS-6 assisted phytoremediation of heavy metals using *Phragmites communis*, Potential application in wastewater treatment. Biores. Technol. 320: 124353.

Shen, W., N. Nada and S. Tachibana. 2000. Involvement of polyamines in the chilling tolerance of cucumber cultivars. Plant Physiol. 124: 431-440.

Shiyab, S. 2019. Morphophysiological effects of chromium in sour orange (*Citrus aurantium* L). Hort. Sci. 54: 829-834.

Singh, H. P., P. Mahajan, S. Kaur, D. R. Batish and R. K. Kohli. 2013. Chromium toxicity and tolerance in plants. Environ. Chem. Lett. 11: 229-254.

Sosnowski, J., E. Malinowska, K. Zankowski, J. I. Kro and P. Redzik. 2019. An estimation of the effects of synthetic auxin and cytokinin and the time of their application on some morphological and physiological characteristics of *Medicago* x *Varia* T. Martyn. Saudi J. Biol. Sci. 26: 66-73.

Srivastava, B. K. and R. P. Singh. 1980. Morpho-physiological response of garden pea (*Pisum sativum* L.) to sowing dates. II-Growth analysis. Ind. J. Hort. 8: 382-389.

Sun, Y. F., J. M. Liang, J. Ye and W. Y. Zhu. 1999. Cultivation of super-high yielding rice plants. China Rice. 5: 38-39.

Tardieu, F., C. Granier and B. Muller. 1999. Modelling leaf expansion in a fluctuating environment: Are changes in specific leaf area a consequence of changes in expansion rate? New Phyt. 143: 33-44.

Tesfaye, K., S. Walkerb and M. Tsubob. 2006. Radiation interception and radiation use efficiency of three grain legumes under water deficit conditions in a semi-arid environment. Europ. J. Agron. 25: 60-70.

Thakur, D. S. and S. R. Patel. 1998. Growth and sink potential of rice as influenced by the split application of potassium with FYM in inceptisols of eastern central India. J. Pot. Res. 14: 73-77.

Van Assche, F., C. Cardinaels and H. Clijsters. 1988. Induction of enzyme capacity in plants as a result of heavy metal toxicity, dose-response relations in *Phaseolus vulgaris* L., treated with zinc and cadmium. Environ. Poll. 52: 103-115.

Vernay, P., C. Gauthier-Moussard and A. Hitmi. 2007. Interaction of bioaccumulation of heavy metal chromium with water relation, mineral nutrition and photosynthesis in developed leaves of *Lolium perenne* L. Chemosph. 68: 1563-1575.

Vyroubalova, S., K. Vaclavikova, V. TureCkova, O. Novak, M. Smehilova, T. Hluska, L. Ohnoutkova, I. Frebort and P. Galuszka. 2009. Characterization of new maize genes putatively involved in cytokinin metabolism and their expression during osmotic stress in relation to cytokinin levels. Plant Physiol. 151: 433-447.

Wang, S., Y. Zhang, W. Ju, J. M. Chen, P. Ciais, A. Cescatti, J. Sardans, I. A. Janssens, M. Wu, and J.A. Berry. 2020. Recent global decline of CO_2 fertilization effects on vegetation photosynthesis. Science. 370: 1295-1300.

West, C., G. E. Briggs and F. Kidd. 1920. Methods and significant relations in the quantitative analysis of plant growth. New Phytol. 19: 200-207.

WHO, 2020. Chromium in Drinking-water (No. WHO/HEP/ECH/WSH/2020.3). World Health Organization, Geneva, Switzerland.

Wilson, W. J. 1981. Analysis of growth, photosynthesis and light interception for single plant stand. Ann. Bot. 48: 507-512.

Wright, I. J., P. B. Reich, M. Westoby, D. D. Ackerly, Z. Baruch, F. Bongers, J. Cavender-Bares, T. Chapin, J. H. C. Cornelissen, M. Diemer, J. Flexas, E. Garnier, P. K. Groom, J. Gulias, K. Hikosaka, B. B. Lamont, T. Lee, W. Lee, C. Lusk, J. J. Midgley, M. L. Navas, U. Niinemets, J. Oleksyn, N. Osada, H. Poorter, P. Poot, L. Prior, V. I. Pyankov, C. Roumet, S. C. Thomas, M. G. Tjoelker, E. J. Veneklaas and R. Villar. 2004. The worldwide leaf economics spectrum. Nature. 428: 821.

Xu, Z. R., M. L. Cai, S. H. Chen, X. Y. Huang, F. J. Zhao and P. Wang. 2021. High-affinity sulfate transporter sultr1;2 Is a major transporter for cr(VI) uptake in plants. Environ. Sci. Technol. 55: 1576-1584.

Yu, Y., W. W. Zhou, K. J. Zhou, W. J. Liu, X. Liang, Y. Chen, D. S. Sun and X. Y. Lin. 2018. Polyamines modulate aluminum-induced oxidative stress differently by inducing or reducing H_2O_2 production in wheat. Chemosph. 212: 645-653.

Yu, Y., W. W. Zhou, X. Liang, K. J. Zhou and X. Y. Lin. 2019. Increased bound putrescine accumulation contributes to the maintenance of antioxidant enzymes and higher aluminum tolerance in wheat. Environ. Poll. 252: 941-949.

Zaheer, I. E., S. Ali, M. H. Saleem, I. Muhammad, G. H. S. Alnusairi, M. A. Bamah, R. Muhammad, A. Zuhaib, M. Rizwan and M. H. Suleman. 2020a. Role of iron-lysine on morpho-physiological traits and combating chromium toxicity in rapeseed (*Brassica napus* L.) plants irrigated with different levels of tannery wastewater. Plant Physiol. Biochem. 155: 70-84.

Zaheer, I. E., S. Ali, M. H. Saleem, M. Ali, M. Riaz, S. Javed, A. Sehar, Z. Abbas, M. Rizwan, M. A. El-Sheikh and M. N. Alyemeni. 2020b. Interactive role of zinc and iron lysine on *Spinacia oleracea* L. growth, photosynthesis and antioxidant capacity irrigated with tannery waste water. Physiol. Mol. Biol. Plants. 26: 2435-2452.