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ORIGINAL ARTICLES

Se-acquisition and reactive oxygen species role in growth, photosynthesis, photosynthetic pigments, and biochemical changes in essential oil(s) monoterpene of Geranium (*Pelargonium graveolens* L.Her.' ex. Ait.)

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ABSTRACT

Se is an essential element required for various metabolic pathways and act as an antioxidant in the various redox-reactions of primary and secondary plant production of – biomolecules. Geranium is an important essential monoterpene oil(s) bearing plant. Culturing the plant at different doses of Se from 0-1.0 mg Se ml^{-1} revealed that Se plays an important role as an in antioxidant promoter, apart from its micronutrient essentiality. 0.25 mg Se ml^{-1} is the critical concentrations for maximum content of (0.21%) total essential monoterpene oil(s). At concentration below and above 0.25 mg Se ml^{-1}, the CO_2 assimilation rate, photosynthetic pigments content and ultimately the accumulation of essential monoterpene oil(s) are affected. The maximum peroxidase and SOD activities were obtained at 0.25 mg Se ml^{-1}, with the production of biomolecule geraniol. Results revealed an oxido-reducable reaction of Se in the formation of monoterpene essential oil(s) and possibly for the major constituents Geraniol.

Key words: Se-acquisition, reactive oxygen species, photosynthesis, essential oil(s) monoterpene, *Pelargonium graveolens.*

Introduction

Selenium is a trace element that can function as an essential nutrient for humans and animals or as an environmental toxicant; the boundary between the two is narrow and depends on its chemical form, concentration, and other environmentally regulating variables (Fan *et al*., 2002; Shardendu *et al*., 2003). Further, it is an important microelement, exists in small amounts in microorganisms, plants, animals and humans. Although selenium is an essential trace nutrient important to humans and most other animals as an antioxidant, toxicity occurs at high concentrations due to replacement of sulphur with selenium in amino acids resulting in incorrect folding of the protein and consequently nonfunctional proteins and enzymes. Distribution of selenium and its species in plants .It was shown, that pea was good accumulators of selenium. The selenium content of pea seeds obtained from the untreated (UT group), once (OT) and twice (TT) foliarly treated plants was determined. The selenium content of pea seeds obtained from the untreated, once and twice foliarly treated plants was, in each case, directly proportional to the number of spraying applications. Seeds are usually a moderate source of selenium, but several studies dealing with cereal and legume seeds showed, that they are able to accumulate high amounts of selenium (Stadlober *et al*., 2001; Smrkolj *et al*., 2005; Smrkolj *et al*., 2006a). Higher selenium contents were found in leaves than in stems of plants grown from both OT and TT seeds, and these contents were, as for the seeds, directly proportional to the number of original foliar

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treatments (Smrkolj *et al*., 2006a). Selenomethionine was the only selenium compound found in supernatants by anion and cation exchange chromatography, comprising 49 and 67 % of the total selenium content in the OT and TT groups, respectively (Smrkolj *et al*., 2006a). The results of the study show that selenium enriched pea seeds are potential source of dietary selenium, on account of their ability to accumulate Se, and that this selenium is present mainly as SeMet, known to be favourable for human consumption (Smrkolj *et al*., 2006a). Common (*Fagopyrum esculentum*) and tartary (*Fagopyrum tataricum*) buckwheat was treated by spraying the leaves with a water solution containing in the form of sodium selenate in the flowering period. The selenium content in all parts of plant was found to be less than 200 ng g−1 in non-treated and in the range 2700–4650 ng g−1 in selenium treated buckwheat. Exposure to UV-B radiation lead to higher selenium accumulation in flowers of both selenium enriched cultivars. In flowers and leaves, on average 11% of the selenium content was soluble and in the form of Se(VI), representing between 0.6% (flowers) and 3% (leaves) of the selenium content. Light stress can promote an accumulation of active oxygen species(AOS) in the chloroplasts in situation where the antioxidant capacity to detoxify AOS is exceeded At low temperature the risk of over –excitation of photosynthesis and formation of AOS is increased as many energy –requiring processes are slowed down and the balance between energy supply and demand is lost resulting in reduction of oxygen (4-6). Antioxidant capacity increases during cold acclimation in several plants as an adaptive mechanism to low temoerature . enhanced antioxidant capacity is also evident during acclination to photoinhibition indicating the important role of AOS scavenging capacity for preventing the injury to the photosynthetic apparatus(9). Selenium is an essential micronutrient needed in antioxidation and hormone balance in lings(10-12). Recent findings show that Se is toxic at higher concentrations , it can exert beneficial effects on plants at low concentrations. Se can increase the tolerance of plants to UV – induced oxidative stress as well as delay in senescence and promote the growth of aging(13-15). The antiaging effect was related to decreased lipid peroxidationand enhanced glutathione peroxidase activity in Se-treated plants. In plants grown at higher light intensities Se counteracted senenscence-related oxidative stress and maintain the green leaf colour longer. Se also decrease the activity of Superoxide dismutase and lowered the amount of tocopherols . Se also promotes H2O2 scavenging by increasing the glutathione peroxidase activity and by the side enhances spontaneous disproportion of super oxide radicals to H2O2 and thus decreases the need for high superoxide dismutase activity(15). One approach to improve the crop plant tolerance to environmental stresses is to increase their antioxidant capacity (16.17). The most recent studies on Zea mays have shown that elevated antioxidant levels can protect the photosynthetic apparatus from oxidative damage.(18,19).In Geranium which is to be cold loving plant improved photosynthetic capacity and productivity at low temperature could be achieved by enhancing the detoxification of of AOS. Study was undertaken to determine whether Se contributes to recovery of photosynthesis fron prolonged high light stress in Geranium. The hypothesis was that through its antioxidative effect Se can alleviate oxidative stress in chloroplass. Further, the response of Geranium to Se supplementation were investigated by monitoring chlorophyll formation its fluorescence and the transcription of antioxidative enzymes. In heavy manuring - N,P,K and excessive vermiculturing as an organic fertigation to increase the crop productivity and sustainability leads, to the major environmental hazardous and limiting factors in crop production . The general response of plants to increase the ion concentration , includes osmotic stress, specific ion toxicity and further the nutrient deficiencies, thus affecting a range of physiological processes involved in cell metabolism(Munns,2002)Further at higher altitudes cultivation . the low pH and high Fe toxicity, apart from heavy metal toxicity due to vermiculturing in plains affects the productivity and biomass of the plants. These cumulative effects of salts complexes and heavy metal in roots , including the other environmental stresses , are known to be mediated atleast partially , by an advocative enhanced generation of Reactive Oxygen Species (ROS)and free radicals. ROS such as hydrogen peroxide (H_2O_2), are generated from the autoxidation of lipids and H_2O_2 production through water-water cycle (Asada 1999, Havaux *et al*. 2003). Leaf antioxidant systems include the enzymes and antioxidant metabolites of the so called water-water cycle, wchich functions to scavange superoxide that is produced from the reduction of oxygen by PSI (Asada,1999). In the water-water cycle, superoxide dismutase(SOD) catalyses the conversion of superoxide to hydrogen peroxide,which is then converted to water via the enzyme ascorbate peroxidase using the ascorbate as a reductant. Formations of these ROS by strong sun shine and UV irradiations arelikely to damage several cellular components such as lipids,proteins, nucleic acid and DNAs through oxidation (Singh 1989, Sawa 2003). Plants contain a wide variety of chemicals that have potent antioxidants – B-carotein, L-tocopherol, ascorbates, Vit.E, Cr, Zn, and Se metals which effects overall metabolism and physiology of the plants through antioxidants activities in scavenging the free-radicals formed in stresses caused by intrinsic ,extrinsic biotic and abiotic factors. Stresses causes a severe imbalence between light absorption and its utilization via photosynthesis . In such conditions , plants emply photoreceptive mechanisms to deal with excess light absorbed by chlrophyll .Two key mechanisms operates: one Xanthophyll cycle-dependent thermal energy dissipation of excess light within the light-harvesting complexes; and second leaf antioxidants enzymes and

metabolites which scavanges free radicals and dangerous ROS that are produced in excess stressesand in the presence of excess light.(Verhoeven *et al*. 2005).Stresses of light harvesting system of higher light illumination light intensity involves major increased in xanthphyll – cycle dependent energy dissipation utilizes xanthophylls carotenoids in a presence of high light energy absorbed by chlorophyll is converted to heat and dissipated harmlessly , thus preventing the formation of singlet oxygen within the light harvesting complexes (Deoimming – Adams and Adms , 1996). Thus, increases include increased pool of xanthophylls – cycle pigments during stresses of light and thus increased in the allocation of absorbed light towards energy dissipation.

Under stresses leaf antioxidants system include the enzyme and antioxidants and its metabolites of the so called warer-water cycle , which scavange superoxides that is produced from thr reduction of oxygen by PS1 (Asada, 1999). Here, the superoxide dismutase(SOD) catalyzes the conversion of superoxide to hydrogen peroxide(H_2O_2), which then converted to water via ascorbate peroxidases using the ascorbate as a reductants . Excess H_2O_2 further affects carbon reduction through Calvin cycle(Takeda *et al*.1995) in chloroplast. L-tocopherol is a lipid – soluble antioxidant metabolite that exists in thalakoid membrane and functions in the detoxification of lipid radicals formed in the presence of singlet oxygen(Havaux, 2003).

Stimulating effects to the additions of small amount of Se on plant growth and yield have been observed by several researchers (Adel M Zayed and Terry, 2003; Terry, 1981). No conclusive evidence yet of an essential role of Se in plant metabolisms but to effect the growth and productivity of plants at low concentration utilization (Huffman and Allaway,1973), either by solublization of Fe^{+3} to Fe^{+2} active Fe for plant uptake or in in an antioxidant activities for defence mechanismsin controlling ROS. Therfore the acquisition of Se on growth and physiology of Geranium with antioxidant activities will be studied in detail.

Geranium, *Pelargonium graveolens* L. `Her, ex Ait. Syn., P. roseum Willd of family Geraniaceae is the only source of one of the most important essential monoterpene oil(s) called the oil of geranium. It is commonly known as rose geranium (Anonymous, 1966, 1975, 1982 and Erickson, 1976). It is distinctly different from the horticultural Geranium, which are basically the ornamental and have no commercial usage in perfumery industries (Douglas, 1969). *P. graveolens* widely grown varieties are Algerian/Funition, Kelkar/Egyption and Bourbon/Reunion (Rajeshwar Rao and Bhatacharya 1992) is popularly cultivated in India. Steam distillation of the shoot biomass of Geranium yields the geraniol and citronellol rich monoterpene oil(s), extensively used for perfuming soaps, and cosmetics to which it imparts a pronounced and lasting rose odour, for high grade perfumes. It is largely used in flavoring tobacco products, tooth pastes, ointments and other pharmaceutical preparations (Ranade, 1988).

Essential oil biosynthesis in Geranium is strongly influenced by Se-acquisition and the stresses caused by Se on nutrition and growth. Se is involved in carbon assimilation and saccharide accumulation, free radical removal, in antioxidant enzymes and role in carbon utilized in terpene biosynthesis and the overall growth of the plants. Furthermore, the role of Se in influencing essential monoterpene oil(s) accumulation seems particularly important. The requirement of Zn antioxidant for Japanese mint and Pelargonium and its limitations imposed on photosynthetic carbon metabolism and translocation in relation to essential oil accumulation in mint have been documented by Misra and Sharma (1991) and Misra *et al*. (2004), respectively. While Se as an antioxidants enzymes for free radical quenching in Geranium have not fully been documented and not much work has been done so far.

In the present paper we report on the role of – Se, as a stimulant of free-radicals quenching through Se affected antioxidants enzyme activity, in Geranium subjected to Se stress. Simultaneously, photosynthetic efficiency in terms of changes in CO_2 exchange rate, photosynthetic pigments, leaf fresh mass, dry mass, leaf area, Se content in whole plant shoot biomass and oil yield were also determined.

Materials and Methods

Plant

Plant tips (5-6 inches) with 3-4 leaves of Geranium (*Pelargonium graveolens*) of Bourbon genotype were obtained from the farm nursery of the CIMAP, Lucknow, India. Uniform cuttings were initially planted in 10000 cm^3 earthen pots filled with purified silica sand (Agarwala and Sharma, 1961), for the development of roots. After 15 days, rooted cuttings were transferred to 2500 cm^3 pots. The salts used in nutrient solution culture were purified for Zn (Hewitt, 1952). Hoagland and Arnon's (1959) nutrient solution was used in the experiment except Fe which was supplied as Fe-EDTA. Three pots each of Zn treatments ranging from 0.0 to 1.0 mg Zn ml^{-1} were maintained in controlled glass house condition at ambient temperature (30 ± 5^oC) and irradiance (between 800-1000 m mol $m^{-}2$ $s^{-}1$). The nutrient solution in each treatment was added at alternate days. With onset of deficiency and toxicity (after 20 days) growth observation and detailed physiological and biochemical data with growth attributes were performed.

CO_2 exchange rate (P_N)

CO_2 exchange rate was measured using a computerized portable photosynthesis system (Srivastava and Misra, 1991) (Model LiCOR 6000, LiCOR, USA).

Chlorophyll (Chl) content

A known mass of leaf tissue (3rd leaf) was extracted with 80% acetone and observance was recorded on spectrophotometer (Pye Unicham PU8610, USA) for determination of Chl a and b according to Arnon (1949)., and Carotenoids were calculated as described by Deming-Adams(1992).

Growth attributes and Zn analysis

Leaf fresh and shoot dry mass and area (area meter LICOR Li-3000) were recorded. For tissue elemental analysis 1.g. dried leaf samples were digested with IN HCl at 60ºC for 24 h. Aliquot samples of the clear digest were diluted with water (10 cm^3) and analyzed for Zn by atomic absorption spectrophotometer (Pye Unicam SP 2800) (Misra and Sharma 1991). Antioxidant reactive peroxidase enzyme activity was estimated as described earlier (Shanon *et al*., 1960). Using 2 g of fresh chopped leaves at position 3rd, were homogenized with 5 ml of 0.1 M phosphate buffer (pH 6.8). Each treatment was replicated 3 times and assayed on SDS page electrophoresis. Superoxide Dimutase (SOD) activity were assayed by the method of Henary et.al (1976).

Estimation of essential monoterpene oil(s)

Geranium oil estimation was done by steam distillation of 100 g freshly plucked leaves in a clevenger's apparatus (Clevenger, 1928). The oil constituents mainly Geraniol, citronellol and other associated oil contents were determined by Gas liquid chromatography (Perkin –Elemer model 3920 B). The stainless steel column was packed with 10% carbowax (20 mesh) on chromosorb WNAW. Injector and detector temperature was maintained at 200ºC. The flow of H_2 was 0.47 cm S^{-1} data processing for area % was done on a Hewllet Packard integrator model HP-33%.

Statistical Analysis

The results were statistically analyzed for the Least Significant Differences (LSD) using the layout of a complete randomized design (CRD). Further the results were analyzed for the correlation coefficient to determine the relationship among the characters studied, using the relationship Y= a+b x.

Results and Discussion

The fresh and dry biomass increased with increase in the level of Zn. (Table 1). Maximum fresh (282.5 g/plant), dry biomass (19.36 g/plant) and leaf area (40.3cm2) was observed at the 0.25 mg Zn ml^{-1} supply. Growth attributes such as, plant height (64.1 cm) was maximum at 0.50 mg Zn ml^{-1} supply. Zn levels of 1.0 mg Zn ml^{-1} were toxic to overall growth parameters. The total chlorophyll content increased upto 0.25 µgZn/ml and then decreased; where as same trend was observed for Chl.a and Chl.b. While Carotenoids were increased from deficient levels of Cr supply to 0.25 mg Zn ml^{-1} supply. The maximum photosynthetic efficiency in terms of CO2 exchange rate of 0.82 µgCO2/m2/s was found at 0.25 mg Zn ml^{-1} supply. The saccharide formation was also found to be highest at 0.25 mg Zn ml^{-1} supply. Zn deficiency and Zn toxicity inhibits P_N in cotton (Ohki, 1976), peppermint (Srivastava *et al*., 1997), Soybean (Ohki, 1978) and sweet mint (Misra *et al* 2004). A decrease in Chl. photosynthetic pigment content represents a decline in photochemical capacity of leaf at deficient Zn supply. (Ohki, 1976).

Peroxidase activity and peroxidase isoenzymes maximum bands were observed at 0.25 mg Zn ml^{-1} cultured plants. The Zn deficient and toxic cultured plants revealed lesser peroxidase activity with lesser peroxidase isoenzymes band profiles. SOD activity showed as Nitro Blue Tetrazollium (NBT) reduction.The maximum SOD (0.248 units/mg.fr.wt.) was obtained at 0.25 mg Zn ml^{-1} cultured plants. In Japanese mint similar report were observed for Mn nutrition (Misra 1996). The maximum monoterpene oil(s) was found at 0.25 mg Zn ml^{-1} treatments. However with respect to oil composition, contents of citronellol. Geraniol, linalool and nerol varied at different Zn treatments (Table 3).As a result of differential Zn supply the content of other micronutrient as Fe, Mn, Zn, and Cu were affected and showed lesser Fe, Mn, Zn and Cu concentrations in shoot tissue of Geranium (Table 3). The maximum (Zn 164 mg g^{-1}), Cu (12 mg g^{-1}) Mn (98 mg g^{-1} and Fe (537 mg g^{-1}) contents were observed at 0.25 mg Zn ml^{-1} treatments.

Table 1: Effect of Se acquisition on growth parameters of Geranium.

Treatment	Plant height (cm)	No. of branches/ plant (Nos.)	Fresh weight /plant (g)	Dry weight/ plant (g)	Leaf area $(cm)^2$
0.0 mg Se ml^{-1}	57.0	9	218.8	14.11	8.2
0.005 mg Se ml^{-1}	58.0	10*	238.6*	16.33*	12.1*
0.05 mg Se ml^{-1}	61.0*	13**	224.8	16.81*	25.2**
0.10 mg Se ml^{-1}	62.5**	18**	252.1**	17.37**	39.1**
0.25 mg Se ml^{-1}	63.4**	10*	282.5**	19.36**	40.3**
0.50 mg Se ml^{-1}	64.1**	10*	215.5	18.46**	37.2**
1.00 mg Se ml^{-1}	59	8	196.2	15.85	11.2
LSD at 5%	2.5	1.1	11.1	2.1	3.5
1%	4.1	3.2	16.3	3.3	6.2

* ** Values are significantly different at 5% and 1% level.

Table 2: Effect of Se on photosynthetic pigments, photosynthesis (P_N), chlorophyll (Chl) contents and saccharide formation of Geranium.

Treatment	Chl a mg g^{-1} f.wt.	Chl b mg g^{-1} f.wt.	Chl a+b mg g^{-1} f.wt.	Caroten-oids mg g^{-1} f.wt.	P_N µg $(CO2)$ m^{-2} s^{-1}	Saccharides µg (CH_2O) m^{-2} s^{-1}
0.0 mg Se ml^{-1}	0.68	0.50	1.18	0.086**	0.15	0.102
0.005 mg Se ml^{-1}	0.79*	0.56	1.35**	0.051**	0.19*	0.129
0.05 mg Se ml^{-1}	0.94**	0.61*	1.55**	0.024**	0.75**	0.510 **
0.10 mg Se ml^{-1}	1.35**	0.69**	2.04**	0.016**	0.76**	0.516**
0.25 mg Se ml^{-1}	1.48**	0.79**	2.27**	0.007**	0.82**	0.558**
0.50 mg Se ml^{-1}	1.01**	0.40	1.41**	0.003*	0.71**	0.483**
1.00 mg Se ml^{-1}	0.82*	0.29	1.11	0.002	0.42**	0.286*
LSD at 5%	0.11	0.08	0.07	0.0002	0.03	0.05
1%	0.15	0.12	0.11	0.0005	0.06	0.07

* ** Values are significantly different at 1% and 5% level.

Table 3: Effect of Se acquisition on geranium total oil and oil composition.

Treatments	% of oil	Citronellol % of total oil	Geraniol% of total oil	Linalool % of total oil	Nerol% of total oil
0.0 mg Se ml^{-1}	0.15	0.21	0.09	8.0	1.1
0.005 mg Se ml^{-1}	0.16	0.27**	0.09	10.0**	1.2**
0.05 mg Se ml^{-1}	0.17*	0.29**	0.10**	9.0**	1.1
0.10 mg Se ml^{-1}	0.19	0.32**	0.11**	6.0**	1.4**
0.25 mg Se ml^{-1}	0.21**	0.25**	0.07**	7.0**	1.2**
0.50 mg Se ml^{-1}	0.16	0.18**	0.12**	8.0**	0.9**
1.00 mg Se ml^{-1}	0.15	0.17**	0.10**	7.0**	0.7**
LSD at 5%	0.02	0.01	0.005	0.04	0.01
1%	0.04	0.02	0.01	0.07	0.02

* ** Values are significantly different at 5% and 1% level.

Table 4: Effect of Se acquisition on tissue concentrations in shoots of Geranium.

Treatments	Fe (mg g^{-1})	Mn (mg g^{-1})	Zn (mg g^{-1})	Cu (mg g^{-1})
0.0 mg Se ml^{-1}	98	26	12	7
0.005 mg Se ml^{-1}	112	37**	19*	9
0.05 mg Se ml^{-1}	142**	41**	34**	11*
0.10 mg Se ml^{-1}	249**	57**	45**	11
0.25 mg Se ml^{-1}	537**	98**	64**	12**
0.50 mg Se ml^{-1}	419**	62**	41**	7
1.00 mg Se ml^{-1}	312**	53**	36**	5
LSD at 5%	21	9	7	3
1%	42	11	9	5

** Values are significantly different at 5% and 1% level.

Statistical analysis showed a positive significant association between Zn content in leaf and total CO_2 exchange rate (P_N) (g=0.924 $\leq$ p=0..5%) and between P_N and saccharides (sugars) (g=0.879 $\leq$ p=0.05% content .However, Zn content in leaf was negatively correlated with Chl. a/b ratio. The CER showed a positive significant association with leaf fresh mass (g = 791 $\leq$ p=0.05%), leaf dry mass (g=692 $\leq$ 0.05%), leaf area and total monoterpene oil(s) (g=0.721 $\leq$ p=0.01). A positive significant correlation was also observed between saccharides and total oil (g=0.695 $\leq$ p=0.01%). A quadratic trend was observed for all these characters which were comparable in + Zn then the deficient and much higher Zn cultured plants.

The observed maximum changes in CER, photosynthesis pigments content, leaf area, and antioxidant enzymes- peroxidase activities along with other leaf physiological parameters at 0.25 mg Zn ml^{-1} supply indicate that this level is optimum for best growth. At this Zn supply activity of peroxidase and its isoenzymes ,the highest CER ,chlorophyll content, and saccharides results in maximum flow of photosynthates to the biosynthetic pathways. The main sites of ROS production in the plant cells are the organelles with highly oxidizing metabolic activities or with sustained electron flows: chloroplasts,mitrochondria and

peroxisomes. (Garnczarska *et al*. 2004). In chloroplasts ROS can be generated by the direct transfer of the excitation energy , or by oxygen reduction in the Mehler reaction (Meloni *et al*. 2003).In addition , H_2O_2 and O_2- may interact in the presence of certain ions (Co, Al, Cu, Cd.Zn, Se) and Cr chelates to produce highly reactive OH (Sudhakar *et al*. 2001). In chloroplasts, H_2O_2 is a powerful inhibitor of the calvin's cycle. Moreover , all ROS can easily react , causing lipid peroxidation and protein denaturation (Yu and Rengel, 1999)therefore the comon effect of free radicals are the degradation of the cell membranes(Prochazkova *et al*. 2001).To prevent such damages plant cells are equipped with an antioxidative system consisting of the low molecular weight antioxidants such as ascorbate, l-tocopherol , glutathione,Xanthophyll carotenoids and protective enzymes (Donahue *et al*.1997) and certain essential Zn,Se. and Cr antioxidant elements(Chakmak, 1999).. Thus during the stress metabolic activities , chain reactions resulting from increase in ROS are less terminated the action of SOD,Fe, Mn. Zn and Cu cofactor mediated enzymes, which are major scavenger of singlet oxygen resulting in the formation of H_2O_2 and O_2.(Meloni *et al*.2003).Less H_2O_2 is then detoxified in the ascorbate – glutathione cycle which involves the oxidation and re-reduction of ascorbate peroxidase and glutathione reductase action, to make water – water cycle(Donahue *et al*. 1997).Antioxidants is thus oxido-reducible processes(Misra, 2003). In healthy unstreesed conditions to the plants not to damage the cell walls and normal physiological activities persists but during the biotic and abiotic stresses of extrinsic factors leads to the antioxidant and antioxidant oxidizing metabolites . Further closing of the stomata and stomatol resistance, for utilization of very low concentration of carbon dioxide which enables then to assimilates carbon dioxide even during considerable stomatol closure(El Bassam, 1998). Thus the study was to examine the stresses of envirnment as well the extrinsic biotic factors to examine simultaneously both ROS of Xanthophyll cycle of carotenoids for dependent energy dissipation and leaf antioxidant system – specially elemental antioxidant chromium(Cr) acquisition in Geranium and its essential monoterpene oil(s) secondary plant products. The simultaneous reduction in growth, CER content of other micronutrient ,and peroxidase activity in leaves indicate the limited availability of photosynthates to essential metabolism and biosynthesis. Utilization of metabolites from primary photosynthetic process into secondary metabolites regulates monoterpene productions (Gershenzon and Croteau, 1991). Thus a close relation between photosynthesis, photorespiaration and terpenoid synthesis also exists in essential monoterpene oil(s) bearing plants (Maffei and Codignola 1990). Moreover, the actively growing leaves requires a larger supply of an antioxidants stimulator Zn, in associations with greater supply of photosynthates and since essential oil biosynthesis occurs in these rapidly growing leaves, the initial growth period would require a still greater supply of
photosynthates and energy .

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