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Research Article

Activity of enzyme Ascorbate peroxidase and catalase in the tissues of roots and stems Azargol and CMS 19 in sunflower- agricultural soil contaminated with heavy metal cadmium

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ABSTRACT

One of the major sources of heavy metal environmental pollutants such as soils that accumulate in the soil and absorbed by plants enters the food chain, of poisoning the plants or causing people feeding them. Heavy metals such as zinc, cadmium, lead can naturally through the weathering of stones and minerals in the soil and accumulate in soil forming process. Cadmium in the periodic table with the atomic number 48 cd is shown. Cadmium on cell division and growth, general plant growth, cell division and plant growth regulation also affects the meristematic region. In order to evaluate the effect of cadmium in sunflower two Azargol and CMS19 was selected. The research was conducted as a factorial randomized complete block design with four replications, with treatments in a greenhouse environment of cadmium in 3 levels (0 - 50 - 100 kg) 3 kg in polyethylene pots with clay and soil the leaves were mixed. According research results ascorbate peroxidase and catalase enzyme activity in the tissues of roots and shoots of sunflower plant figures showed The highest enzyme activity of ascorbate peroxidase in root tissue in Azargol and CMS 19, and the lowest activity in the stem tissue as well as the highest catalase activity in the root and the lowest stem tissue was seen in the treatment of 100 ppm cadmium . As a result Azargol with the activity of APX at the root of their resistance and catalase has any activity against high concentrations of cadmium in no way CMS19 the stress of heavy metals in high concentrations of ascorbate peroxidase activity but compared with Azargol showed resistance to cadmium stress of resistance is slower.

Key words: Sunflower, APX, Cadmium, Catalase, Resistant

INTRODUCTION

One of the major sources of heavy metal environmental pollutants such as soils that accumulate in the soil and absorbed by plants enters the food chain, poisoning the plants or causing people feeding them [3].Heavy metals such as zinc, cadmium, lead and can naturally through the weathering of rocks and minerals in the soil and accumulate in soil forming process but this natural resource in pollution from human activity, including construction industrial, mining, fossil fuel, chemical fertilizer use and organic industrial waste and sewage sludge are not comparable[22]. The annual world production of large amounts of sewage sludge is

a significant amount of farmland used as fertilizer in the presence of toxic elements such micronutrients as well as lead, cadmium, mercury and nickel in the ground Agriculture, agricultural limit[4]. Heavy metals and toxic metals such as cadmium and lead as necessary to stimulate metabolism and the creation of free radicals and reactive oxygen species were considered [18]. As well as some heavy metals such as cadmium, lead and mercury at high concentrations, on the growth and yield of plants affected. [7][23]Cadmium in the periodic table with the atomic number 48 cd is shown. Elemental relatively rare, soft, bluish-white and part of the transition metals [31] The element cadmium in the industry as an anti-friction, anti-color alloy used in industry as well as in the semiconductor, nuclear reactors, metalworking, ceramics, installations p. v. c, plastics industry, production of batteries, pesticides, fungicides, motor oil, car tires and photography applications. In the field of cadmium as fertilizer or lime materials contaminated with heavy metals cadmium and cadmium concentrations in soil used has increased from 5 to 14 times [1]. Cadmium can be found in the main organs of the human body such as the kidneys and lungs accumulate and lead to some dangerous diseases such as cancer [2] of cadmium on the division and growth of cells, the overall growth of the plant, meristematic zone of cell division and also influence plant growth regulation it. [12] and as a toxic metal known that high concentrations cause oxidative stress. [11] Excessive amounts of cadmium may decrease nutrient uptake, inhibition of enzyme activity, changes in enzymes of the antioxidant defense system [32]. Studies show that plants such as sunflower (Helianthus annuus L.) high tolerance to heavy metals [6, 34 and 39] and sunflower oilseed crop is considered one of the scientific name and the name of the English sunflower Helianthus annus in the world. The origin of this plant is North America and the largest area under sunflower cultivation quit as Russia, Ukraine, India and Argentina. Cytoplasmic male sterile hybrid seeds also can be produced by Azaftabgrdan. [13] In fact, among the 67 species of this genus are known today, there are many perennial species, about 17 species of them can be considered as domesticated sunflowers are largely decorative and often difficult to distinguish is them. One of the most important plants for the climate of our country are sunflower seed oil with high quality and relatively high tolerance to drought and water stress our country has played an important role in agriculture. [16] The origin of the plant tolerance to heavy metals in the plant various mechanisms in the plant cell wall, and Non antioxidant glutathione precursor an important and valuable for the purification plant is the key role in the plant is responsible for the disposal of metal poisoning.[5] And the experimental results showed that a series of plant enzymes sensitive to metals can be 10 to 1000 times the concentration of cadmium in the form of complex with pc compared to ion free radicals, endure. [17] Of defense mechanism against oxidative stress from free radicals include: 1. preventive mechanisms, 2-chemical mechanism, 3-physical mechanisms, 4-antioxidant defense mechanisms [29] Some plants to defend against free radicals (ROS) and antioxidant activities such as catalase, superoxide dismutase, glutathione peroxidase to wash and wash and clean up free radicals are used. [9] [26] One of the main causes of damage to the plants of the plant is exposed to environmental stress for plants cope with environmental stress and oxidative activity of antioxidant enzymes start, such as superoxide dismutase (SOD), ascorbate peroxidase (APOX) glutathione reductase and (GR), peroxidase (POX), catalase (CAT) and antioxidants such as ascorbic acid and glutathione are low molecular weight [15] The effects of cadmium in response antioxidant activities such as: (SOD, CAT, APOX, MAD, POX) and the exchange of sister chromatids in

the form of beans were measured and the results found that SOD, CAT in stressed plants treated with 50 and 100 micro-molar of cadmium were detected and treated with 50 micro-molar APOX activity significantly increased heavy metal cadmium. Antioxidant enzymes superoxide dismutase and catalase, ascorbate peroxidase and glutathione reductase enzymes in plants is very important especially in the first line of defense superoxide dismutase plant has been against free radicals. [36]

MATERIALS AND METHODS

Seeds of sunflower seeds 2 hybrid of resistance and sensitive cultivars (AZARGOL & CMS19) were used and the seeds from the Seed and Plant Improvement Institute, were prepared. Research in 1391 at the Islamic Azad University, Science and Research Laboratory and Agricultural Biotechnology Research Institute of Iran. The project was carried out for every 2 genotype heavy Tymarflz Sunflower 3 levels of cadmium, which include treatments zero, 50 and 100 kg in 4 replicates in a randomized complete block design was a factorial design [24] Plants in the greenhouse with the same temperature and light conditions for 2 months grew, the plants in pots with a mixture of clay and peat PE 3 kg to 5.2 kg in the same proportion from each of Soils were planted in pots and cadmium stress were mixed. Seeds were sown in pots at a height of 3 cm and by controlling temperature and humidity in the greenhouse continued growth Yaft.ps within one month of the symptoms of stress in the plants, began sampling. The root, stem, pot plants and various tissues were separated. Nakano and Asada method for measuring the activity of APX in 1981 [28] Catalase activity by Kerouac and colleagues in 1993 to measure the enzymatic activity of the sunflower plant was used. Samples frozen in liquid nitrogen freezer temperature -80 ° C the tank was placed and tissue samples from the roots and stems of plants can be worn separately in a mortar, then powder samples for extraction. After measuring the enzyme preparation accurate data analysis using statistical program SPSS 18 05/0 was performed at the level of the average measurement indicators were grouped using Duncan test.

RESULTS AND DISCUSSION

Enzyme activity of catalase (CAT) in Azargol and CMS 19

Based on analysis of variance 1-1 catalase activity in root and shoot the interaction between treatment and genotype is significant in 01/0 level that can be 99% sure that catalase activity is likely to treatment used and depends on the genotype used.

(**Table 1-1**)Analysis of variance catalase activity in tissues of the stem and root Azargol and CMS19 in sunflower

		Mean square (MS)	
S.O.V	**df**	stems catalase activity	Root catalase activity
Repeat	3	0/0000002418	**0/0000002392**
Treatment	1	0/000001367^{ns}	**0/0000005882^{ns}**
Genotype	1	0/000**	**0/003****
G*T	1	0/001**	**0/000****
Error	9	0/0000002875	**0/0000001822**
Total	15	–	**–**

** Significant at the level 0/01. * Significant at level 0/01 .ns no significant difference statistically

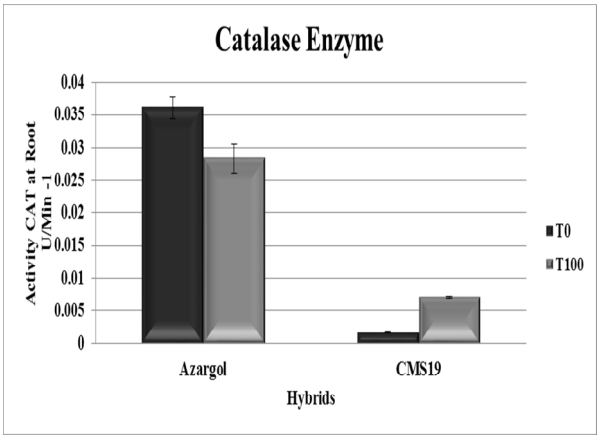
1-2 reviews enzyme catalase (CAT) in root tissue Azargol and CMS19

Based on analysis of variance 1-1 catalase activity in root organ interaction between

treatment and genotype is significant in 0.01 level that can be stated with 99% probability ensure that the treatment and catalase activity used depends on the genotype.

Figure 1-2 enzyme activity of catalase (CAT) in root organ Azargol concentration 0ppm (control) was most active in the territory and about the amount of 0/036 U/min $^{-1}$ and low catalase activity in this cultivar 0/ 028 U/min $^{-1}$, which is almost equal to more than cadmium concentration control and the 0/001 U/min $^{-1}$ is. The results showed that the enzyme activity of catalase (CAT) in the concentration of cadmium absorbed by plant roots, nothing to do with the accumulation of cadmium in the roots, but catalase in the CMS19 minimal stress but Azargol Activity CMS19 was better and probably another defense mechanism to cope with the heavy elements there and it was found that the mechanism of resistance activity of the enzyme catalase (CAT) at the root of the CMS19 effect.

Research Schutzen Double et al in 2001 [33] showed that cadmium causes oxidative stress in pea plant roots and also reduce the heavy metal cadmium in catalase activity by pine tree roots and leaves of sunflower, [21] and leaf rice [15] and the leaves of Arabidopsis [10] and radish roots [37] and the leaves of soybean [34] observed.



(Fig 1-1**)** comparison chart of catalase activity in root tissue Azargol and CMS19 in sunflower

2.1 reviews enzyme catalase (CAT) in stem tissue Azargol and CMS19

According to Table 1.1 Analysis of variance enzyme activity of the enzyme catalase (CAT) in stem tissue interaction effect between treatment and genotype is significant in 0/01 level that can be 99% sure that catalase activity is likely to and the treatment used depends on the genotype used.

Research continues on Figure 1-3 Active enzyme catalase (CAT) 100ppm concentration of cadmium in the soil in the straw Azargol happened that almost the amount of 0/032 U / min^{-1} and enzyme activity in the CMS19 in similar concentrations of heavy metal cadmium is almost equivalent to a 0/012 U / min $^{-1}$ in second place in the investigation and proceed with the activity of catalase in Azargol and CMS19 the concentration 0ppm (control), respectively 0/017 U / min^{-1} and 0/023 U / min $^{-1}$ CMS19 more about the activities of Azargol showed.

The results showed that the enzyme activity of catalase (CAT) in stem tissue in more than CMS19 Azargol in terms of tension, but in normal conditions and stress-free catalase activity was observed in more CMS19. Catalase activity decreased with increasing concentrations of cadmium in some plants due to the reduction in plant proteins in the metal toxicity and oxidative stress has been reported [38]

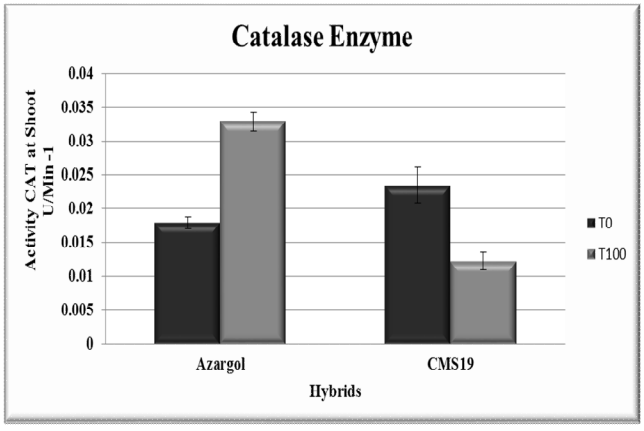


Fig 3.1 Comparison Chart of catalase activity in the straw Azargol and CMS19 in sunflower

3-1 enzyme ascorbate peroxidase (APX) root and shoot Azargol and CMS19

Based on the analysis of variance table 2-1 ascorbate peroxidase enzyme activity in root and shoot the interaction between treatment and genotype is significant in 0/01 level is likely to be 99% sure ascorbate peroxidase enzyme used as treatment and also depends on the genotype used.\

Table 1-2 Analysis of variance ascorbate peroxidase activity in different organs and CMS19 Azargol in sunflower

s.o.v	df	Mean square (MS)	
		Ascorbate peroxidase activity stem	Ascorbate peroxidase activity Root
Repeat	**3**	**0/000**	**0/001**
Treatment	**1**	**0/017****	**0/009****
Genotype	**1**	**0/0000001140**^{ns}	**0/007****
G*T	**1**	**0/003****	**0/020****
Error	**9**	**0/000**	**0/000008839**
Total	**15**	_	_

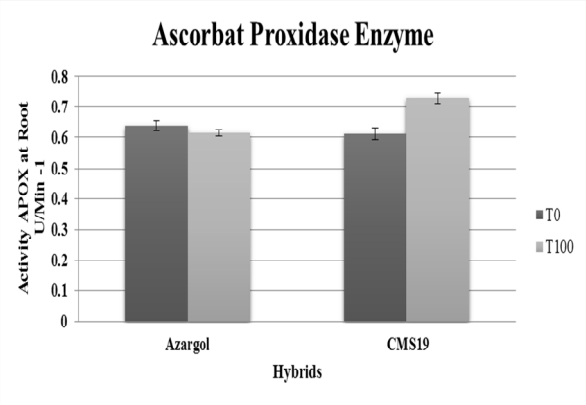
** Significant at the level 0/01. * Significant at level 0/01 .ns no significant difference statistically

2.2 study the activity of the enzyme ascorbate peroxidase (APX) in root tissue Azargol and CMS19

According to the Analysis of variance table 2-1 ascorbate peroxidase enzyme activity in root organ interaction between treatment and genotype is significant in 0/01 level that can ensure 99% probability ascorbate peroxidase enzyme used as treatment and the time depends on the genotype used.

Figure 2-2 activity ascorbate peroxidase enzyme (APOX) CMS19 genotype in concentration of 100 ppm in the soil in the root organs most active and almost equal to the amount of 0/172 U / min^{-1} and also the lowest figure in the same enzymatic activity and at low concentrations of cadmium in the soil is almost equivalent to a 0/61 U / min^{-1} and the investigation of the activity of APX Azargol zero and 100ppm concentrations to a level almost 0/162 U / min^{-1} and the results showed that the enzyme activity of ascorbate peroxidase (APOX) relationship with the concentration of cadmium absorbed by plant roots, and found that the mechanism of resistance in parts of ascorbate peroxidase enzyme activity in the plant root no effect. Kumchai et al., 2013 [19] to research in resistance of the Capitata canola, rapeseed Capitata than the cadmium, molybdenum and zinc were analyzed and the results were reported total anthocyanin content in the plant against heavy metals cadmium and zinc 1 and 0.1 mm, respectively, at concentrations higher than the other treatments micron and micron and less Li at a concentration of 5 mg also increased the content of anthocyanins in plants and thus the activity of antioxidant enzymes such as: CAT, APOX, SOD at high concentrations of heavy metals such as cadmium, molybdenum and start their activities. Gross et al in 2011 [27] on the species of plant called Ginseng Glomerata the studies, the effects of different concentrations of cadmium treated with Mqadyr0-45- 90 mol per liter observed that the activity of the enzyme superoxide dismutase (SOD) in leaf in the early days and in the roots after 20 days increased and this, along with increasing concentrations of cadmium is, the stress of cadmium enzyme activity APOX in the leaves increased while root activity APOX at the concentration of cadmium reduction is. The researchers reported that radish and pea shoots and roots of plants under stress, heavy metal cadmium, enzyme activities, CAT, SOD, GR, APOX, DHAR increases. [20] [30] and also Grvpa and colleagues reported in 2001

that the plants under stress to respond to toxic heavy metals such as zinc and antioxidant enzyme activities of CAT, SOD, APOX increase. [14]



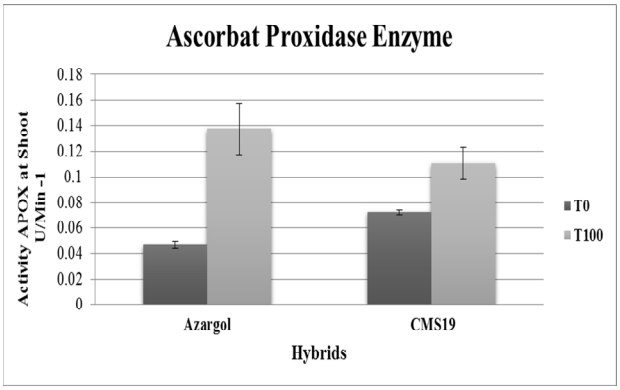
(Fig 2-2) Comparison Chart of ascorbate peroxidase enzyme activity in the body and CMS19 in sunflower root Azargol

2-3 Study enzyme ascorbate peroxidase (APX) in stem tissue Azargol and CMS19

Based on the Analysis of variance table 2-1 ascorbate peroxidase enzyme activity in stem tissue interaction effect between treatment and genotype is significant in 0/01 level is likely to be 99% sure ascorbate peroxidase enzyme used as treatment and the used depends on the genotype.

The Figure 2-3 following topics highest enzyme activity ascorbate peroxidase (APOX) at 100ppm concentration of cadmium in the soil straw Azargol in the amount equal to the 0/13 U / min $^{-1}$ and enzyme activity in the CMS19 the concentration of approximately 0/11 U / min $^{-1}$ is ranked second, followed by examining the activity of APX in Azargol and CMS19 the control concentration (0 ppm) was almost the same amount and the same enzymatic activity showed.

The results showed that the enzyme activity of ascorbate peroxidase (APX) related to the concentration of cadmium uptake in organs not shoot as well as the resistance mechanism in the plant stem does not affect the activity of APX and possibly by other mechanisms of defense against the figures of stress your heavy elements of support. In research conducted by Unyuyal and colleagues reported in 2010 that antioxidant enzymes CAT, APX, SOD, GR important role in protecting species of bean plants exposed to toxic heavy metal cadmium plays. [36]



(Fig 2-3) Comparison Chart of ascorbate peroxidase enzyme activity in the body and CMS19 in sunflower stem Azargol

REFERENCES

1. Hoseinian, M. 1370. Heavy metals and environmental pollution. Journal of Water. My number.
2. Falamaki Amin Hussein Tavalloli, M. Eskandari and M. Moradi Estahbanati, improvement of soils contaminated with cadmium and copper by using di-calcium phosphate, water and soil conservation magazine, Issue I, Fall 1392, ISSN: *2251-7*
3. Antoniadis, N. and Alloway, B. J. 2001. *Availability of Cd, Ni and Zn to rye grass in seawage sludge treated soils at different temperatures*. water, Air and soil pollut. 132: 201 – 204.
4. Adamu,C. A. Bell, P. F. Mulchi, C. and Chaney, R. 1989. *Residual metal concentration in tobacco a decade following farmland application of municipal sludge*. J . Environ. pollut. 56 : 113 –126.
5. Abdul R.Memon and Peter Schroder.(2009).”*Implications of metal accumulation mechanisms to*

phytoremediation".Environ sci pollut Res.16:162-175.

6. Bennett LE, Burkhead JL, Hale KL, Terry N, Pilon M, Pilon-Smits EAH (2003) *Analysis of transgenic Indian Mustard plants for phytoremediation of metal contaminated mine tailings.* J Environ Qual 32:432-440
7. Baker, A. J. M. and Brooks, R. R. (1989) *Terrestrial higher plants which hyperaccumulate metallic elements. a review of their distribution, ecology and phytochemistry*. Biorecovery 1: 81-126.
8. Brusko, I., Smith, E. C. and Murphy, S. D. 2005. *Interference of soybean cultivars with Alopecurus muosuroides*. Weed Sci. 49: 85-92.
9. Ben Amor N., Ben Hamed K., Debez A., Grignon C., Abdelly C. 2005. *Physiological and antioxidant responses of the perennial halophyte Crithmum maritimum to salinity*. Plant Sci. 168, 889-899.
10. Choo K.S., Snoeijs P., Marianne Pedersén M., 2003, *Oxidative stress tolerance in the filamentous green algae Cladophora glomerata and Enteromorpha ahlneriana*, J. Exp. Mar. Biol. Ecol.,298: 111–123
11. Cho UH, Park JO.2000. *Mercury-induced oxidative stress in tomato seedlings*. Plant science 156,1-9
12. Das P, Samantaray S & Rout GR (1997). S*tudies on cadmium toxicity in plants*: A review. Environmental Pollution 98: 29–36.
13. Fernandez-Martinez, J.M. B.perez-vich,L.*velasco*,*sunflowe*r,chapter 6,1995
14. Groppa M. D, Tomaro, M. L. and Benavides, M. P . 2001. "*Polyamines asprotector against cadmium or copper-induced oxidative damage in sunflower leaf discs*," Plant Sci., vol. 161, pp. 481-488, Aug.
15. KUO.M.Ch., and KAO.Ch.H.,2004. *Antioxidant enzyme activities are upregulated in response to cadmium in sensitive, but not in tolerant, rice (Oryza sativa L.)*seedlings. Bot. Bull . Acad–.Sin.) 45:291-299.
16. karimzadeh-Asl, K.H. D. Mazaheri, and S.A. Peyghambari. 2003. *Effect of four irrigation intervals on the seed yield and quantitative characteristics of three sunflower cultivars. Iranian Journal of Agricultural Science. 34(2): 293-301. (In Persian).*
17. Kneer R, Zenk MH (1992). *Phytochelatins protect plant enzymes from heavy Metal.* poisoning.- phytochemistry. 31: 2663-2667.
18. Kaznina N.M., Laidinen G.F., Titov A.F., Talanov A.V. 2005. *Effect of lead on the photosynthetic apparatus of annual grasses*. Izv. Akad. Nauk Ser. Biol. 2: 184-188.
19. Kumchai, J. Huang, J. ZLee, . C. YChen, . F. C. and Chin. S. W. 2013. *The Induction of Antioxidant Enzyme Activities in Cabbage Seedlings by Heavy Metal Stress.* World Academy of Science, Engineering an Technology 73.pp:465-470
20. Lombi, E. Zhao F. J., Dunhan, S. J. and McGrath, S. P. (2000) "*Cadmium accumulation in populations of Thlaspi caerulescens and Thlaspi goesingense*," New Phytol., vol. 145, pp.11-20, Jan..
21. Laspina N.V., Groppa M.D., Tomaro M.L. & Benavides M.P. (2005) *Nitric oxide protects sunflower leaves against Cd-induced oxidative stress*. Plant Science 169, 323–330.
22. Mauskar J.M. 2007. *Cadmium –An Environment Toxicant, Central Pollution Control Board, Ministry ofEnvironment & Forests, Govt of India*, Parivesh Bhawan, East Arjun Nagar,Delhi-110032.
23. Madhava Rao. K. V. and Sresty, T. V. S. 2000. *Antioxidative parameters in the seedling of pigeonpea in response to Zn and Ni stresses*.plant Sci. 157: 113 – 128 .
24. Moteshare-Zadeh b, Savaghebi-Firozabadi Gh R, Alikhani H A, Mirseyed-Hosseini H

(2008).*Effect of Sunflower and Amaranthus Culture and Application of Inoculants on Phytoremediation of the Soils Contaminated with Cadmium*. American-Eurasian J. Agric. & Environ. Sci., 4 (1): 93-103. ISSN 1818-6769

25. Memon, A.R. and Schroder, P., 2009. *Metal accumulation in plants and its implication in phytoremediation*. Environmental Science and Pollution Research International, 16(2): 162-175.
26. Mittova V., Tal M., Volokita M., Guy M. 2003. *Up-regulation of the leaf mitochondrial and peroxisomal antioxidative systems in response to salt-induced oxidative stress in the wild salt-tolerant tomato species Lycopersicon pennellii*. Plant Cell Environ. 2: 845-856.
27. McGrath, S. P. Micó, C. Curdy, R. and Zhao, F. J. 2010. "*Predicting molybdenum toxicity to higher plants: Influence of soil properties*," Environ. Poll., vol.158, pp. 3095-3102, Oct..
28. Nakano Y., Asada K. 1981. *Hydrogen peroxide is scavenged by ascorbate-specific peroxidase in spinach chloroplast*. Plant Cell Physiol. 22: 867-880.
29. Przybyl K., Wo!ny A. 2004. *Komórka w warunkach stresu "rodowiskowego*. t 2. Wyd.UAM, Pozna, 172 pp.
30. Romero-Puertas M, Corpas FJ, Rodriguez-Serrano M et al. *Di`erential expression and regulation of antioxidative enzymes by cadmium in pea plants*. J of Plant Phys 164: 1346-1357, 2007.
31. Stormier, Friedrich ;*Role of Cadmium in the improvement of human* life.1835. Engineering Science
32. Sandalio L.M., Dalurzo H.C., Gómez M., Romero-Puertas M.C. & del Río L.A. (2001) *Cadmium-induced changes in the growth and oxidative metabolism of pea plants*. Journal of Experimental Botany 52, 2115–2126
33. Schützendübel A., Schwanz P., Terchmann T., Grossk Langeenfeld-Heyger R., Godbold D.L. & Polle A. (2001) *Cadmiuminduce changes in antioxidative systems, hydrogen peroxide content, and differentiation in Scots pine roots*. Plant Physiology 75, 887–898.
34. Sobkowiak R, Rymer K, Rucińska R, Deckert J (2004) *Cadmium- induced changes in antioxidant enzymes in suspension culture of soybean cells*. Acta Biochim Polon 51:219–222.
35. Tang, S., Kishore, V.K. and Knapp, S.J. (2003a) *PCR-multiplexes for a genome-wide framework of simple sequence repeat marker loci in cultivated sunflower*. Theor. Appl. Genet. 107, 6–19.
36. UNYAYAR1.S., GUZEL DEĞER1.A., CELİK1.A., CEKİC2.F.O., CEVİK1.S.,2010. *Cadmium-induced antioxidant status and sister-chromatid exchanges in Vicia faba L*. Turk J Biol 34 : 413-422.
37. Viktória A.P., Lea P.J. & Azevedo R.A. (2001*) Antioxidant enzymes responses to cadmium in radish tissues*. Phytochemistry 57, 701–710.
38. Vajpaee, P. R D. Tripathi, U. N. Rai, , M. B.Ali and Singh, S. N. 2000. *Cd Accumulation reduces chlorophyll biosynthesis, Nitrate reductase activity and protein content in Nymphaea alba L*. Chemosphere. 41: 1075 –1082.
39. Wu YS, Tang KX. *MAP Kinase cascades responding to envi- ronmental stress in plants*. Acta Bot Sin. 2004; 46 (2): 127-136