G.J.B.B., VOL.1 (1) 2012: 104-109 ISSN 2278 – 9103

GLOBAL JOURNAL OF BIO-SCIENCE & BIOTECHNOLOGY

www.scienceandnature.org

PHYTOREMEDIATION POTENTIAL OF *CASSIA TORA* (L.) ROXB. TO REMOVE HEAVY METALS FROM WASTE SOIL COLLECT FROM SUKALI COMPOST AND LANDFILL DEPOT, AMRAVATI (M.S.).

Shirbhate Nayana & Malode S. N.
P.G. Department of Botany,
Govt. Vidarbha Institute of Science and Humanities, Amravati – 444 604
Email:-satishmalode17@gmail.com

ABSTRACT

The present study was undertaken to find out the problems and prospects of Municipal solid waste and to study various physicochemical parameters and heavy metals of soil which is collect from Sukali compost and landfill depot, Amravati. This paper is aim to investigate the capacity of *Cassia tora* (Caesalpinioideae) to remove contaminant in waste soil polluted by various heavy metals. *Cassia tora* endemic to India is a potential to hyper accumulator of heavy metals. During 2 month experiment the plants of *Cassia tora* grown in control and waste soil. Quality of waste soil have been studied with respect to important physicochemical parameters such as pH, soil colour, texture, temperature, conductivity, Cl, Na, K, etc. Pollution of waste soil is controlled by various methods by using certain plants and microbes. Due to favourable conditions for growth of *Cassia tora* plants has recorded higher total chlorophyll content in waste soil (0.88 mg chl. /g) than control (0.86 mg chl./g) after 30 days and after 60 days waste soil (1.71 mg chl./g) than control (1.23 mg chl./g). T.S. of stem showed accumulation of certain pollutants from waste soil, its effect on secondary xylem tissue and break the tissue of pith region. All recorded differences in morphological, anatomical and chlorophyll content of plants were caused by differences of cumulative environmental conditions with dominant effects of the contamination degree of locations of soil. Heavy metal analysis of initial waste soil, it contained Cu (0.748 mg/g), Zn (5.032 mg/g), Cr (0.220 mg/g), Ni (0.492 mg/g), Fe (18.90 mg/g), Mn (6.081 mg/g), Co (-0.157 mg/g) these all metal concentrations lower down except Zn, Cr and Fe after the *Cassia tora* grown in this soil during 2 month period and Cu (0.581 mg/g), Zn (0.184 mg/g), Cr (0.465 mg/g), Ni (0.1005 mg/g), Fe (22.09mg/g), Mn (4.811 mg/g), Co (-0.022 mg/g). If this Municipal solid waste landfill continues, it may create a serious environmental problems, present investigation study will revealed that *Cassia tora* have a capacity to accumulate certain metals from waste soil and analyzed the amount of toxicity and various nutrients of soil.

KEY WORDS: *Cassia tora*, phytoremediation, Sukali compost and landfill depot, Toxicity, Pollutants, Heavy metals.

INTRODUCTION

Human evolution has lead to immense scientific and technological progress. Global development, however, raises new challenges, especially in the field of environmental protection and conservation (Bennett *et al*., 2003). Nearly every government around the world advocates for an environment free from harmful contamination for their citizens. However, the demand for a country's economic, agricultural and industrial developments outweighs the demand for a safe, pure, and natural environment. "Phytoremediation is the use of plant to remove, degrade harmless hazardous materials present in the soil or groundwater". This emerging technology may offer a cost effective, nonintrusive and safe alternative to conventional soil clean up technique by using the ability of certain tree, shrub, herb and grass species to remove, degrade or immobilize harmful chemicals from the soil (Chaney *et al*., 1997). Therefore, the present study was undertaken to find out the problems and prospects of Municipals solid waste and to study various physicochemical parameters and heavy metals of soil which is collect from Sukali compost depot, Amravati.

Amravati city has a population of 6, 78,192 in 2008 including 1, 28,682 rural and 5, 59,510 urban persons. Amravati is a city in the state of Maharashtra, India and seventh most populous metropolitan area in Maharashtra. According to Health Department, Amravati Municipal Corporation, Amravati report of 2009-2010, around 692.11 TPD solid wastes is generated every day (Personal Communication with Dr. Jadhav). The total area of Sukali composed depot of about 9.5 hectors, it totally polluted due to solid waste. The plants which grow on the solid waste materials are mostly influenced by the local flora and some of them are exotic. They may be helpful for solid waste management and also reduce environmental hazards by absorbing organic matters that appeared from decaying of some solid wastes. *Cassia* is a legume in the sub family Caesalpinioideae. It grows wild in most of the tropics and is considered a weed in many places. Many researchers stated that some plants belongs to genus *Cassia* accumulate heavy metals in their tissues. Siringoringo, (2000) reported that *Cassia multijuga* was found to be highly capable of absorbing and accumulating Pb.

MATERIALS AND METHODS

Selection of site and sample collection:- Soil samples were collected; in summer season 2009-2011 from 3 different spots of Sukali compost depot **(Plate1)** and three pits of 20-50 cm depth were dug for soil sampling at selected points.

.

PLATE 1. Experimental site of sukali compost and landfill depot, Amravati (M.S.)

Collection and germination of Seed: - *Cassia tora* seeds were collect from different sites of Campus of Government Vidarbha Institute of Science and Humanities (G. V. I. S. H.) Amravati in the year 2009. Seeds were germinated in control soil collected from Botanical garden, P.G. Department of Botany, G. V. I. S. H., Amravati and waste soil from Sukali compost and landfill depot. Each bag contains 5 kg of soil sample. Total 25 seeds sown in each bag and % of germination were counted **(Plate 2).**

PLATE 2: Cassia tora(L.) Roxb. Grown in experimental field trial

Physicochemical analysis: - Moisture content (Dhyansingh *et al.*, 1999) and soil texture **(**Arora and Pathak, 1989) was analyze. pH, Electrical conductivity, and temperature of the soil was measured in a 1:2 or 1:5 soil- water suspension or in saturates soil paste. Colour notations indicated by using Munsell`s soil colour chart.

Mineral and salt analysis: - Na, K and Ca ions were determined by flame photometer (Hanway and Heidel, 1952). The organic carbon in the sample was oxidized with potassium dichromate and sulphuric acid (Walkely and Black, 1934). Calcium carbonate by titrimetric method (Piper, 1966)**.**The chloride content of the soil was directly measured by titrimetric method (Santra *et al*., 2006).

Metal analysis: - Detection and analysis of metal ions such as Cu, Zn, Cr, Ni, Fe, Mn and Co, from soil and sediments, wet oxidation of sample was carried out. Wet oxidation employs oxidizing acids like HNO_3 –$HCLO_4$ di-acid mixture (Jackson, 1958).

Effect of pollutants on anatomy:-Thin sections were stained with saffranin and light green, observed and photographed using inverted microscope (Axiovert 40c) for detail study of cell structure.

Effect of pollutants on chlorophyll contents of plants: - Chlorophyll was extracted in 80% acetone and absorbance at 663 nm and 645 nm was read using UV-Visible spectrophotometer (Elico SL 164). Leaves samples with control and treatments were estimated following the procedure of Whatley and Arnon (1963).

Statistical analysis: - On the three replications, a completely randomized block design was used in the experiment. The difference between control and waste soil compared using *t* tests, and significant differences were found at ($p< 0.01$), ($p< 0.05$), ($p< 0.2$).

RESULTS AND DISCUSSION

Growth: - Maximum (88 %) germination was found in waste soil than control soil 51% **(Table 1)** because of the favourable condition available in waste soil. The waste soil collected had a sandy texture together with the following parameters pH-7.76, moisture content-3.93%, Organic carbon- 43.17 %, Chlorides - 49.7 mg/kg, Na-26.5 mg/kg, K- 89 mg/kg, $CaCO_3$-79.1 %.The plants of each pots were harvested after 30 - 60 days the maximum biomass production was observed in waste soil 12.57-28.76 gm it was 50-60% higher over the control 5.82-12.02gm it was totally due to various nutrients in greater quantity in waste soil, it show stimulating effect on the growth and biomass production**.** According to Anikwe and Nwobodo (2001), Municipal wastes increase the nitrogen, pH, cation exchange capacity, percentage of base saturation and organic matter. Organic waste can provide nutrients for increased plant growth, and such positive effect will likely encourage continued land application of these wastes (Anikwe and Nwobodo, 2001; Nyles and Ray, 1999).

Chlorophyll content: The *Cassia tora* plants were grown in waste soil has significantly higher chlorophyll content than control. Relation in total chlorophyll contents after 30 - 60 days the maximum obtained in waste soil 0.88-1.71 mg chl./gm than control 0.86-1.23 mg chl./gm **(Table 1).** However, relation between chlorophyll a and chlorophyll b content after 30-60 days in waste soil samples Chl. a = 0.53-0.80 mg chl. /gm; Chl. b = 0.34-0.90 mg chl./gm and control Chl. a= 0.49-0.75 mg chl./gm; Chl.b =0.36-0.47 mg chl./gm. Della *et al*., (1998) suggest that the variation in chlorophyll content has been used in many studies in order to investigate the effect of pollutants on plants.

TABLE 1: Growth performance of *Cassia tora* in field trial

Parameters	After 30 Days		After 60 Days	
	CS	WS	CS	WS
Seed germination (%)	51	88*	-	-
Biomass(gram)	5.82	12.57***	12.02	28.76*
Total chlorophyll(mgchl./gm tissue)	0.86	0.88***	1.23	1.71*
Chl. a(mg chl./gm tissue)	0.49	0.53***	0.75	0.80**
Chl.b(mg chl./gm tissue)	0.36	0.34	0.47	0.90**
Na (mg/Kg)	0.1	0.2	0.15	0.25
K (mg/Kg)	30	37	31.5	46
Ca (mg/Kg)	97	79	101	84

CS- Control Soil, WS- Waste Soil

NS- Non significant, ***$p < 0.2$, **$p< 0.05$, *$P< 0.01$

Physicochemical analysis of soil

The rise in temperature of soil accelerates chemical reactions, decrease solubility of gases and pH of soil. pH of initial waste soil (pH 7.76) at temperature 32.4^0 C mostly towards the alkaline side and deviated from neutral (pH 7) to alkaline side and it rises slightly after the *Cassia* grown in this soil (pH 8.32) at 31.5^0 C. Colour of soil is useful indicator of some of the general properties of soil, such as the amount and state of organic matter and soil aeration, the colour of control soil is dark reddish brown and waste soil is greyish dark brown, due to water logging. Moisture content in waste soil was found to be very low (3.93 %) as compared to control soil (10.13 %), wetness depends largely on the porosity of a soil, and for that reason clayey soil, which have a high porosity generally have larger water content than do sandy soils. The organic carbon content in initial waste soil (43.17 %) it 30-40 % higher than in control soil (34.35%) after the *Cassia* grown in waste soil it totally absorbed by plant after 2 month, the organic carbon much lowered down . High amount of organic carbon influences the soil physical and chemical properties and it also plays a vital role in soil productivity it enhances intensive root growth which leads to accumulation of organic residues in the soil (Chopra and Kanwar, 2002). The very low organic content of soil may be attributed to the poor vegetation and high rate of organic matter decomposition under hyper thermic temperature regime which leads to extremely high oxidizing condition (Kameriya, 1995). The chlorides content in initial waste soil (49.7 mg/ kg) it was 45-50 % higher than control soil (28.4 mg/kg) after 2 month growth of *Cassia* in waste soil (28.4 mg/kg) it lowered down and equal to control. The conductivity of initial waste soil was much higher ($1.792X10^6$) than control ($0.128\ X10^6$), it become regulate after the *Cassia* grown in waste soil ($0.512\ X10^6$).With regards to Na, K, Ca and $CaCO_3$ content, the initial waste soil contains more amount of the micronutrients than control except Ca which is higher in control soil it indicating the high productivity of the waste soil. In waste soil Na, K and $CaCO_3$ concentration 50-70 % higher than garden soil (control) **(Table 2)**. This followed the pattern reported by Isirimah *et al.,* (2003) for productive agricultural soil. The high base saturation of the dump soil may be as a results of increase release of Na, K, Ca and $CaCO_3$ by decomposing waste, is an indication of good yield.

TABLE 2: Physicochemical analysis of initial control and waste, grown *Cassia tora plant* during 2 month

Sr.No.	Parameters	Initial control soil	Initial waste Soil	Plant grow in Control Soil (CS)	Plant grow in Waste Soil (WS)
1.	Temperature (^{0}C)	25	32.4	29.9	31.5
2.	pH	8.47	7.76	8.26	8.32
3.	Colour	Dark reddish brown	Grayish dark brown	Dark reddish brown	Grayish brown
4.	Moisture content (%)	10.13	3.93	8.54	4.75
5.	Moisture correction factor (mcf)	1.10	0.13	1.08	1.04
6.	Soil texture	Sandy loam	Sandy	Sandy loam	Sandy
7.	Organic Carbon (%)	37.67	43.17	27.82	Nil
8.	Chlorides (mg/Kg)	28.4	49.7	28.4	28.4
9.	Conductivity µmoho/m	0.128 X10^6	1.792X10^6	0.128 X10^6	0.512 X10^6
10.	Na (mg/Kg)	7	26.5	3.5	9.0
11.	K (mg/Kg)	10	89	12	71
12.	Ca (mg/Kg)	700	400	710	490
13.	$CaCO_3$ (%)	22	79.1	19.44	83.2

Phytoremediation study of *Cassia tora* plant

Concentration of Cu, Zn, Cr, Ni, Fe, Mn and Co metal in the waste soil is shown in the **(Table 3).** There is a difference in the concentration of ions in control and waste soil. Concentration of various metal ions in initial control and waste soil was found to be, in waste soil Cu (0.748 mg/g), Zn (5.032 mg/g), Cr (0.220 mg/g), Ni (0.492 mg/g), Fe (18.90 mg/g), Mn (6.081 mg/g) and Co (-0.157 mg/g) these all values are higher except Cu and Fe as compared to control soil, Cu (0.899 mg/g), Zn (2.684 mg/g), Cr (0.168 mg/g), Ni (0.061 mg/g), Fe (22.41 mg/g), Mn (4.695 mg/g) and Co (-0.075 mg/g). Copper content in the initial waste soil was found to be 0.748 mg/g its 35-40% higher over the control it reduced significantly 20-25 % after the *Cassia* grown in waste soil during 2 month 0.581 mg/g from the initial waste soil total 0.222 mg/g Cu absorb or extract or stabilize by the plant. Copper ions form strong coordination complexes with organic matter (Stevenson, 1976; Stevenson, 1991). Reddy (1995) found the proportion of Cu bound to organic matter in the soil solution increased from 37 to 95% as the pH decreased. Concentration of Zn in waste soil (5.032 mg/g) its 50-55 % it's higher than control (2.684 mg/g). *Cassia* sp. absorbs 95-96 % more amount of Zn (4.185 mg/g) from waste soil. Zn present in higher concentrations, it showed stimulating effect on growth and Biomass production. Chromium concentration in waste soil (0.465 mg/g) rises within a 2 month experiment as compared to initial concentration of Cr in waste soil (0.220 mg/g). Findings by Gardea-Torresdey *et al*., (2004) support Cr being concentrated in the roots and not translocated to the aerial parts of the plant by determining the uptake and accumulation of Cr by *Convolvulus arvensis* L. The concentration of Ni from the initial waste soil (0.492 mg/g) its 83-84 % higher over the control, it becomes significantly reduced 79-80 % after 2 month 0.100 mg/g, in this process plant absorb 0.339 mg/g of total Ni ion. High Iron content was found in control soil (22.41 mg/g) as compared to waste soil (18.90 mg/g) after the experiment it become again rise 10-20 %. That is due to the reason that iron is present in various forms of iron oxides in plant as well as soil. Manganese tends to be higher 59-60 % in waste soil (6.081 mg/g) it becomes reduced to 20-21 % within 2 month *Cassia* grown in soil (4.811 mg/g). Complexes of Mn in the soil solution of a sandy loam increased from 10 to 55% as the amount of organic matter in the soil increased (McGrath *et al.,* 1988). Cobalt was totally absent in control as well as in waste soil. Higher concentration of Co it may prove toxic to plant and severely interfere with physiological and biochemical functions (El-Sheekh *et al*., 2003). The observation of growth of the plant over 60 day period it clearly demonstrates that plant can tolerate Cu, Zn, Ni and Mn ions in waste soil condition as the field experiment, the samples were procured from Sukali compost and landfill depot, Amravati.

TABLE 3: Comparative study of metal phytoremediation from control and waste soil through *Cassia tora* plant

Sr.No.	Type of Soil	Metals Ions mg/ g						
		Cu	Zn	Cr	Ni	Fe	Mn	Co
1.	Initial Control Soil (I-CS)	0.899	2.684	0.168	0.061	22.41	4.695	-0.075
2.	Plant absorbs Ions in Control Soil (60 days) (PAM-CS)	0.006	1.710	0.057	0.034	4.959	1.66	-0.156
3.	Total Metals Remains in Control soil (TMR-CS)	0.806	2.200	0.303	0.033	22.24	4.4035	-0.122
4.	Initial Waste Soil (I-WS)	0.748	5.032	0.220	0.492	18.90	6.081	-0.157
5.	Plant absorbs Ions in Waste Soil (60 days) (PAW-WS)	0.222	4.185	0.562	0.339	5.219	1.81	-0.0234
6.	Total Metals Remains in Waste soil (TMA-WS)	0.581	0.184	0.465	0.100	22.09	4.811	-0.022

Anatomical characteristics

There are some specific anatomical changes were observed from the samples of stem from both the treatments (Plate 3). Stem consist of single layer epidermis which bear ridges and furrows, with cuticle, 4-5 layered cortex and vascular system contain secondary xylem and phloem tissues. Secondary xylem contains xylem vessels and medullary rays. Larger xylem cavities found in waste soil growing plant as compared to plants grown in control soil. Secondary phloem 4-5 layered. Anatomy of root showed accumulation of certain nutrients and metals which are beneficial for plant growth, due to more secondary root produced. Plants usually adapt to high pollutant concentrations and unfavourable environmental conditions (Davis and Beckett, 1978; Wyszkowski and Wyszkowska, 2003), which is likely to result in different morphology and anatomy. Well developed cuticle on the surface of leaves originating from that Ada Huja (polluted site) was observed. Also, stomata cavities were larger in leaves of *Tansy* plants from both localities. Leaf mesophyll from Ada Huja (polluted site) consists of 1 or 2 layers of elongated palisade cells. Palisade cell had many chloroplasts and large intercellular cavities. Changes in shape and structure of thin walled mesophyll cells have been widely reported (Stevovic *et al*., 2010).

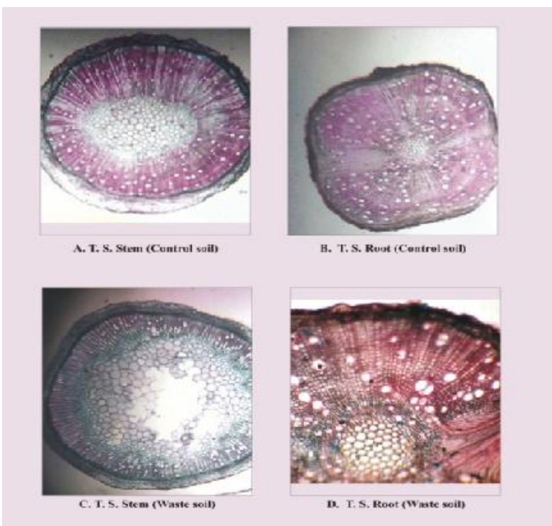

Plate 3. Microscopic study of *Cassia tora*

ACKNOWLEDGEMENT

The authors are extremely thankful to Amravati Municipal Corporation, Amravati, for permitting soil sampling from Sukali compost and landfill depot, Amravati. Authors are also thankful to DST-New Delhi for providing necessary infrastructure facility under FIST programmed, Department of Botany, G. V.I.S.H. Amravati. Authors personally thankful to Dr. Jadhav, Health department, Amravati (M. H.) for providing necessary data related to work.

REFERENCES

Anikwe, M. A. N. and Nwobodo K.C.A. (2001) Long Term Effect of Municipal Waste Disposal on Soil Properties and productivity of Sites used for Urban Agriculture in Abakaliki, Nigeria. Bioresources Technol. 83,241-251.

Arora, S. and Pathak, S. C. (1989) Laboratory techniques in modern biology. 2ndEdi.KalyaniPublishers,New Delhi-110002, 73-89.

Bennett, L. E. , Burkhead, J. L., Hale, K.L., Terry, N., Pilon, M. and Pilon Smits ,E. A. H. (2003) Analysis of transgenic Indian mustard plants for phytoremediation of metal contaminated mine tailings. J. Environ. Qual., 32, 432- 440.

Chaney, R. L., Malik, M., Li, Y. M., Brown, S.L., Brewer, E.P., Angle, J.S. and Baker, A. J. (1997) Phytoremedition of soil metal. Curr Poun Biotechnol 8, 279-284.

Chopra, S. L. and Kanwar, J.S. (2002) Analytical agricultural chemistry. Kalyani publication, New Delhi, India. pp. 1081- 1082.

Davis, R. D., Beckett P. H. (1978) Critical levels of twenty potentially toxic elements in young spring barley. Plant Soil, 49, 395-408.

Della, T., Ferranti, G., Lupattelli, F., Pocceschi, N., Figoli, A., Nali, C. and Lorenzini G. (1998) Effect of ozone on morpho- anatomy and physiology of Hedera helix. Chemosphere. 36,651-656.

Dhyansingh, Chhonkar, P. K., Pandey, R.N. (1999) Soil plant and water analysis –A method manual. IARI, New Delhi.

El-Sheek, M. M., Naggar, A. H. El., Osman, M. E. H. and El-Mazaly, E. (2003) Effect of cobalt and growth pigments and the photosynthetic electron transport in *Monoraphidiumminutum* and *Nitzchiaperminuta*. Braz. J. Plant physio., 15,159.

Gardea-Torresdey, J. L., Peralta-Videa, J. R., Montes, M., De La Rosa, G. And Corral-Diaz, B., (2004) Bioaccumulation of cadmium, chromium and copper by *Convolvulus arvensis* L.: impact on plant growth and uptake of nutritional elements. Bioresource Technology, 92, 229-235.

Hanway, J. J. and Heidel, H. (1952) Soil analysis methods as used in lowa state college soil testing laboratory. Lowa Agri.57, 1-31.

Irishimah, N. O., Igwe, C. and Iwegbue C. M. A., (2003) Important ions in soils Environment In: Introductory Soil Chemistry and Biology for Agric and Biotech. OsiaIn'I Pub. Lt. PH, 34-97.

Jackson, M. L. (1958) Soil and Chemical Analysis.Prentic-Hall, Englewood Cliffs, NJ, USA.

Kameriya, P. R. (1995) Characterization of soil of agro climatic zone of transitional plain of inland drainage zone 11 A, of Rajasthan, Ph. D. Thesis, R.A.U., Bikaner.

McGrath, S. P., Sanders, J. R. and Shalaby, M. H. (1988) The effects of soil organic matter levels on soil solution concentrations and extractabilities of manganese, zinc and copper. Geoderma. 42,177-188.

Nyle, C. B. and Ray, R. N. (1999) The Nature and Properties of Soils. 12th Ed. United States of America, 743-785.

Piper, C.S. (1966) Soil and plant analysis. Hans's publications, Bombay, 224.

Reddy, K. J., Wang, L. and Gloss, S. P. (1995) Solubility and mobility of copper, zinc and lead in acidic environments. Plant and Soil. 171, 53-58.

Santra S.C., Chatterji, T. P. and Das, A. P. (2006) College Botany Practical.Vol.1New Central Book Agency (P) Ltd., 221.

Siringoringa, H. H. (2000) The role of some urban forest plants in adsorbing lead particulates. Bull. Penelitian-Hutan, 622,1-16.

Stevenson, F. J. (1976) Stability constants of Cu^{2+}, Pb^{2+}, and Cd^{2+} complexes with humic acids. Soil Science Society of America Journal. 40, 665-672.

Stevenson, F. J. (1991) Organic matter-micronutrient reactions in soil. In 'Micronutrients in Agriculture'. 2nd. edn. (ed. Mortvedt, J.J., Cox, F.R., Shuman, L.M. and Welch, R.M.) Soil Science Society of America, Madison, 145-186.

Stevovic S., Mikovilovi, V. S. and Dragosavac, D. A. (2010) Environmental impact on morphological and anatomical structure of Tansy. African Journal of Biotechnology Vol. 9 (16), pp. 2413-2421.

Walkely, A. J. and Black, I. A. (1934) Estimation soil organic carbon by chromic acid titration method. Soil sci. 37, 29-38.

Whatley, F. R. and D. I. Arnon (1963) Methods in Enzymology, Academic Press, New York,1, 308.

Wyszkowski, M., Wyszkowska, J. (2003) Effect of soil contamination by copper on the content of macro elements in spring barley. Pol. J. Nat. Sci. 14, 309-320.