Thesis of doctoral (PhD) dissertation

A STUDY ON COPPER-TOLERANCE OF GIANT REED (*Arundo donax* L.) ECOTYPES FOR BIOENERGY PURPOSES

Nevien Adel Ismail Elhawat

Supervisor: Prof. Miklós Fári *D.Sc.*

UNIVERSITY OF DEBRECEN

KERPELY KÁLMÁN DOCTORAL SCHOOL

Debrecen

2015

1. INTRODUCTION AND AIMS OF STUDY

Giant reed displays unique physiological features whereby it readily absorbs and concentrates toxic chemicals from contaminated soil with no appreciable harm to its own growth and development. It is one of the mostly used plants as a trace element bio-accumulator, especially via phytoremediation processes, due to its capacity of absorbing contaminants such as metals that cannot be biodegraded. Giant reed can grow in different environments with spacious ranges of pH, salinity, drought and trace metals without any symptoms of stresses and can easily adapt to different ecological conditions and grow in all types of soils. However, because of its great adaptability to different ecological conditions, giant reed is considered noxious invasive weeds in riparian habitats throughout the world.

Cu is an essential micronutrient that is found in the upper layer of soils. Naturally, the total Cu contents in soils range between 2-109 mg kg^{-1} and world-soil average is 38.9 mg kg^{-1}. Cu is a rather immobile element in soils and shows relatively little variation in total contents of soil profiles. The Cu content is strongly associated with soil texture, where light sandy soils contain less Cu than loamy soils. Soils usually restrain Cu in many forms depending upon soil pH.

The objectives of this study were:

- Assess the possible use of Cu-contaminated soil for purposes of bioenergy production during the phytoremediation process.
- Study the growth dynamics of giant reed ecotypes under elevated Cu concentrations in water bodies as well as in Cu-contaminated soil-like growth medium (sand).
- Determine the threshold of Cu on growth of biomass candidate giant reed.
- Calculate the bioconcentration and transportation factors, bioaccumulation percent, and removal rate of giant reed ecotypes for Cu.
- Evaluate the Cu-tolerance capacity of different giant reed ecotypes originated from epigenetic modifications.
- Measuring the efficiency of giant reed ecotypes to clean Cu-contaminated water bodies.

2. MATERIALS AND METHODS

2.1. Source of plant material

Four ecotypes of giant reed were tested in the present work. All ecotypes had almost similar morphological features.

- Blossom (BL) ecotype was brought from South Carolina, USA.
- 20SZ and STM ecotypes were brought from Újszentmargita village, Hungary (Pro-Team Co. Ltd.).
- ESP ecotype was brought from Spain (Biotek Co Ltd, K-12)

All plant materials used for the current study were somatic embryo-derived plantlets of giant reed (*Arundo donax* L.) according to patent application of the University of South Carolina (Márton and Czakó, 2002). Embryogenic callus culture of the BL ecotype was originally obtained from the University of South Carolina, while other ecotypes were obtained from MOP Biotech Co. Ltd. (Nyíregyháza, Hungary). All ecotypes were propagated in the Ottó Orsós Laboratory, Department of Plant Biotechnology, University of Debrecen, Debrecen, Hungary.

2.2. *In vitro* experiment

An *in vitro* experiment was carried out in order to investigate the efficiency of giant reed ecotypes (BL and 20SZ) grown on MS medium (Murashige and Skoog, 1962) to remove Cu from artificially Cu-contaminated water bodies and monitoring impacts of Cu on growth dynamics of giant reed ecotypes. Two identical size aseptic seedlings (5 cm height) were grown on MS medium (Murashige and Skoog, 1962) supplemented with increasing concentrations of Cu under aseptic conditions as described by Márton and Czakó (2004; 2007).

2.2.1. Experiment layout and growth conditions

The randomized complete block design (RCBD) was used for experiment layout with five replications, each having two plants per tube. The treatments included control (no Cu) and six doses of copper (II) sulfate pentahydrate ($CuSO_4 \cdot 5H_2O$) i.e., 0, 1, 2, 3, 5, 10, and 26.8 mg L^{-1} Cu. Two giant reed plantlets of BL and 20SZ ecotypes almost equal morphological characteristics were directly transplanted to autoclaved glass tubes (2.5 × 25 cm) containing MS medium as described by Márton and Czakó (2004; 2007) for 6 weeks under gnotobiotic conditions. The cultivated tubes were set under white fluorescent lamps (41 µmol m^2 s^{-1} photon flux density), and 16/8 h light/dark cycle and temperatures of 30 ºC during the day and 27 ºC during the night. The average relative humidity was recorded to be 75%.

2.3. *Ex vitro* experiment

A greenhouse experiment was carried out, between July and October 2013, using 1-kg plastic pots in order to assess growth effeciency of three ecotypes of biomass candidate giant reed (BL, STM, and ESP) growing on elevated Cu concentrations. In addition, biomass production of giant reed under high Cu levels was evaluated with aim to determine the possible directing of Cu-contaminated soils towards renewable energy production during the phytoremediation process as additional benefit. A randomized complete block design (RCBD) was used for experimental design with five replications. Two identical (5 cm height) and aseptic seedlings of giant reed ecotypes were transplanted in 1-kg plastic pot.

2.3.1. Experiment installation and treatments

Sand (quartz, SiO_2) was used as growth medium (soil-like growth medium) for this research. Sand was bought from Hungarian market (producer Aquabau Magyarország Kft., seller Buamax Company, Debrecen, Hungary). Sand was washed with diluted HCl and then rinsed many times in distilled water. No nutriets were detected in HCl-washed sample of sand. Each plastic pot was filled up with almost 1 kg of air-dry sand. Copper (II) sulfate pentahydrate ($CuSO_4 \cdot 5H_2O$) was used to prepare the treatments of Cu. Four different levels of Cu were applied, i.e., 100, 200, 300, and 400 mg Cu kg^{-1} against control (no Cu). All pots were received Cu treatments and then irrigated by distilled water to reach its saturation percent and left to the next day to attend equilibrium. In the morning, two plantlets of studied giant reed ecotypes were treansplanted per each pot and then all experimental pots were kept under foliar tent in order to acclimatize the seedlings with greenhouse conditions. During the whole experimental period all pots were kept at its water holding capacity (WHC) (almost 75% of its saturation percent) using distilled water in order to prevent the leakage of Cu from the pot. The following nutrients were added to experimental pots; $(NH_4)_2SO_4$ 10 mg kg^{-1}; $Na_2HPO_4 \cdot 12H_2O$ 10 mg kg^{-1}; K_2HPO_4 10 mg kg^{-1}; $CaCl \cdot 2H_2O$ 2.6 mg kg^{-1}; $MgCl_2 \cdot 6H_2O$ 2 mg kg^{-1}; $FeCl_3 \cdot 6H_2O$ 0.5 mg kg^{-1}; $MnSO_4 \cdot H_2O$ 0.1 mg kg^{-1}. The experiment started in July 2013 and was harvested by beginning of November. Number of new buds, length of plant, and number of leaves were recorded twice, after 6 and 10 weeks from plantation, during the experiment period.

3. RESULTS AND DISCUSSION

3.1. *In vitro* experiment

3.1.1. Cu transport to giant reed plants

Results of Cu concentrations determined in the different parts of the plant and in the growth medium are presented in Fig. 1. At lowest Cu concentration (1 mg L^{-1}), Cu content in root, culm, and leaves was 2.99, 1.50, and 0.84 for BL ecotype and 0.41, 0.16, and 0.19 for 20SZ ecotype. At 26.8 mg L^{-1} treatment, Cu content in root, culm, and leaves of BL plants was 76.57, 24.28, and 23.44 µg g^{-1} DM, while 55.91, 69.49, and 10.18 µg g^{-1} DM were measured in root, culm, and leaves of 20SZ plants, respectively. Moreover, for Cu accumulation in different plant parts, both ecotypes BL and 20SZ showed the same tendency where the order of Cu accumulation was: nutrient solution < leaf blade < culm < roots. The mass balance remained maintained for both ecotypes, which was in agreement with Wu Qi et al. (2012).

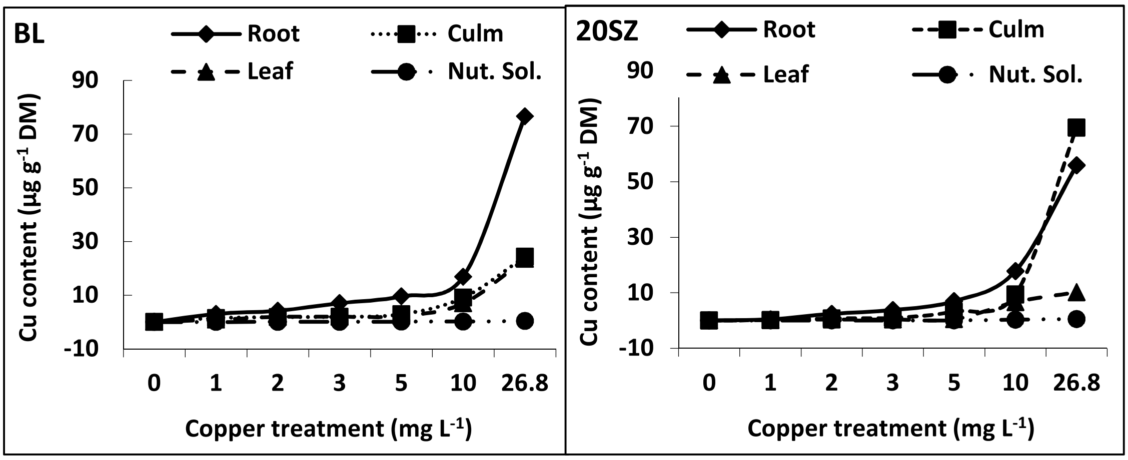

Fig. 1: Cu content in various plant parts of BL and 20SZ ecotypes and nutrient solution after 6 weeks experiment.

3.1.2. Bioconcentration and transportation factors of giant reed ecotypes

The bioconcentration (BCF) and transportation (TF) factors for both BL and 20SZ ecotypes showed high efficiency for concentrate and transport Cu from aqueous solution (Fig. 2a, b). For instance, BCF of BL and 20SZ ecotypes was above 1, where the highest estimated values for BL and 20SZ were 5.34 and 5.06, respectively, for Cu treatment of 1 and 26.8 mg L^{-1}, respectively. On the contrary, the lowest values were

2.93 and 0.76 at 5 and 1 mg L^{-1} treatment, respectively, for BL and 20SZ ecotypes, respectively (Fig. 2a). On the other hand, TF in both ecotypes was almost down 1 except for 20SZ at highest treatment (26.8 mg L^{-1}) since it was 1.43. The highest calculated values for TF were 0.95 and 1.43 at 10 and 26.8 mg L^{-1}, respectively, for BL and 20SZ ecotypes, respectively (Fig. 2b). Similarly, same results for TF of giant reed plants grown on Cu-contaminated soil were documented by Wu Qi et al. (2012).

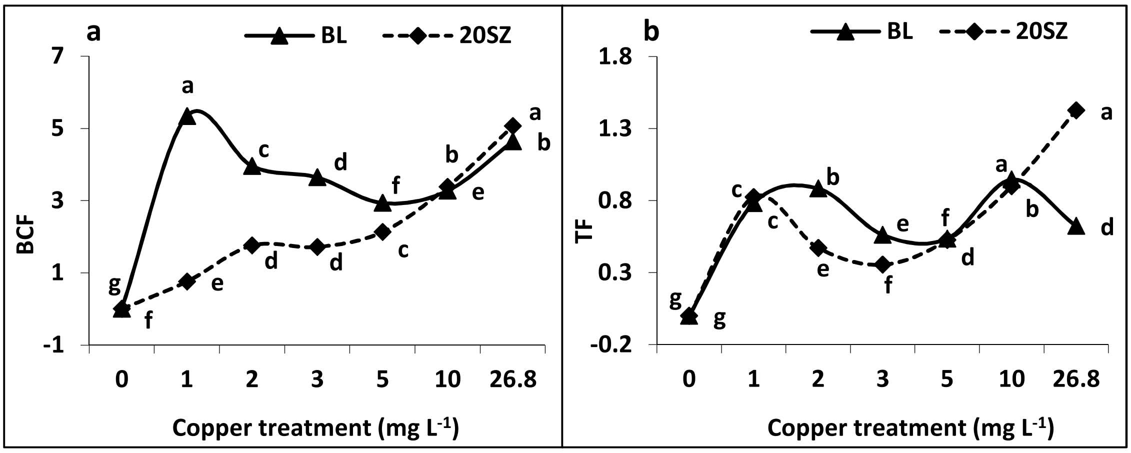

Fig. 2: **a**, Bioconcentration factor (BCF) of BL and 20SZ ecotypes at elevated Cu concentrations. **b**, Transportation factor (TF) of BL and 20SZ ecotypes at different Cu concentrations. Different letters on same line show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

3.1.3. Bioaccumulation percent and removal rate of Cu

Data clearly illustrated Cu was concentrated predominantly in roots (51.4–65.1 %) and culm (17.9–28.2 %) followed by leaves (15.7–24.4 %) for BL ecotype. While 20SZ ecotype was not so far from BL ecotype, where Cu was accumulated in roots (41.2–73.8 %) and culm (18.4–51.3 %) followed by leaves (7.5–19.7 %) (Table 1). In general, by looking at the average values for Cu bioaccumulation percent (BAP) in different plant parts of both ecotypes under different Cu treatment, it can be concluded that almost 59.3 % of Cu uptake by BL ecotype retained in its roots and 28.2 % in culm followed by 12.5 % in leaf blades. Whereas the accumulated Cu in plants of 20SZ ecotype was distributed as follow: 58.6 % in roots, 22.6 % in culm, and 18.9 % in leaf blades. Table 1 showed that the removal rate was the same for both ecotypes and ranged between 96.6 and 98.8 % at 2 and 26.8 mg L^{-1} treatments for BL ecotype. Total removal 100 % was observed with 20SZ ecotype at 1, 2, 3, and 5 mg L^{-1} treatments then slightly decreased with increase Cu concentration in culture medium achieving 97.0 and 98.0 % at 10 and 26.8 mg L^{-1} treatments, respectively.

Table 1: Effect of different Cu concentrations on removal rate (RR) and bioaccumulation percent (BAP) of BL and 20SZ ecotypes

Cu (mg L^{-1})	Bioaccumulation percent (BAP), %						Removal rate (RR), %	
	BL ecotype			20SZ ecotype			BL ecotype	20SZ ecotype
	Root	*Culm*	*Leaf*	*Root*	*Culm*	*Leaf*		
0	0.0g	0.0f	0.0f	0.0g	0.0f	0.0f	0.0g	0.0d
1	56.1d	28.2a	15.7e	54.8d	20.5de	24.8a	98.5b	100.0a
2	53.1e	22.5c	24.4a	68.0b	21.4d	10.6c	96.6f	100.0a
3	64.0b	17.9e	18.0d	73.8a	18.4e	7.8d	97.5d	100.0a
5	65.1a	19.6d	15.3e	65.6c	30.1b	4.4e	97.2e	100.0a
10	51.4f	27.6b	21.0b	52.7e	27.6c	19.7b	98.2c	97.0c
26.8	61.6c	19.5d	18.9c	41.2f	51.3a	7.5d	98.8a	98.0b

In the columns are different letters showing significant differences among each group of treatments according to Duncan's test at $p < 0.05$

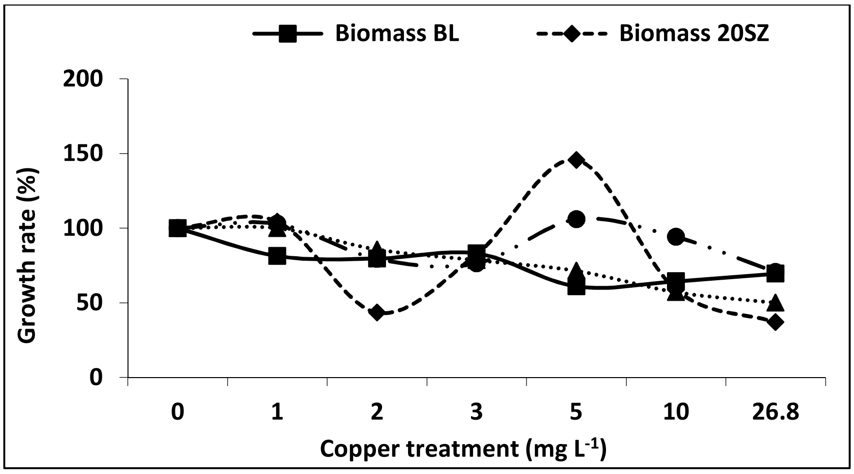

Fig. 3: Effect of Cu concentrations on shoot biomass and culm length of BL and 20SZ ecotypes of giant reed. Growth is expressed as a percentage of the no Cu control.

3.1.4. Cu sensitivity and plant growth

Results obtained showed increasing Cu concentration in the nutrient solution slightly decreased the growth of both ecotypes (Fig. 3). However, BL ecotype was more sensitive for increasing Cu concentration in culture medium than 20SZ ecotype. Where, gradually decreasing in shoot biomass percent compared to control plants was found. At

26.8 mg L^{-1} treatment for BL ecotype, 30 % reduction in shoot biomass was recorded compared to control plant. Otherwise, 20SZ ecotype plants showed more tolerant characters, where 45.7 % increasing in shoot biomass was found at 5 mg L^{-1} treatment, but more than 63 % reduction in shoot biomass was noticed at the highest treatment. The performance of culm length was similar to shoot biomass.

3.2. *Ex vitro* experiment at greenhouse

3.2.1. Dry mass of giant reed ecotypes

BL plant showed negative response towards increasing Cu doses, where with increase added Cu the dry mass of root system gradually reduced till treatment of 300 mg kg^{-1} but increased again at 400 mg Cu kg^{-1} (Fig. 4). Dry mass of BL ecotype decreased from 3.29 to 1.29 g $plant^{-1}$ when Cu concentration changed from 0 to 300 mg kg^{-1}. Moreover, in all treatments up to 400 mg kg^{-1} BL root system had the highest values among all treatments. Unlike BL ecotype, STM and ESP ecotypes had hesitated values for dry mass of root system with elevated Cu doses. For STM ecotype, treatment of 100 mg kg^{-1} of Cu enhanced dry mass of root system, while other treatments had lower values of dry mass compared to control plant. ESP plants recorded the highest dry mass values at treatment of 200 mg kg^{-1}, and lowest measured dry mass of its root system was found at treatment of 400 mg kg^{-1} (Fig. 4).

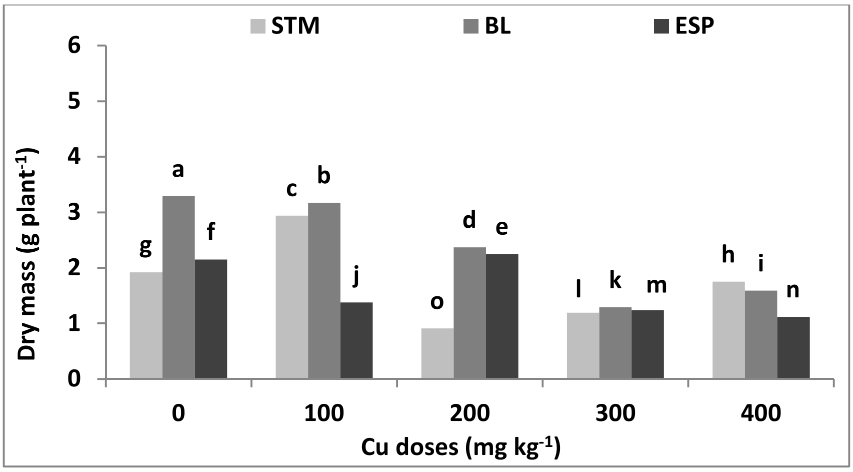

Fig. 4: Dry mass of root system of giant reed ecotypes (STM, BL, and ESP) at harvest after grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Dry mass of culm was positively affected with increasing Cu doses (Fig. 5). All ecotypes had higher values of culm dry mass compared to control plants. With increasing Cu levels in soil-like growth medium dry mass of culm of STM, BL, and ESP increased signficantely. The highest dry mass of BL culm was 5.45 g plant^{-1} and was achieved at treatment of 100 mg kg^{-1}, while highest value of STM culm was 4.81 g plant^{-1} when Cu concentration in soil-like growth medium was 300 mg kg^{-1}. But ESP plants had highest dry mass of culm at treatment of 200 mg Cu kg^{-1} and it recorded 5.20 g plant^{-1} (Fig. 5).

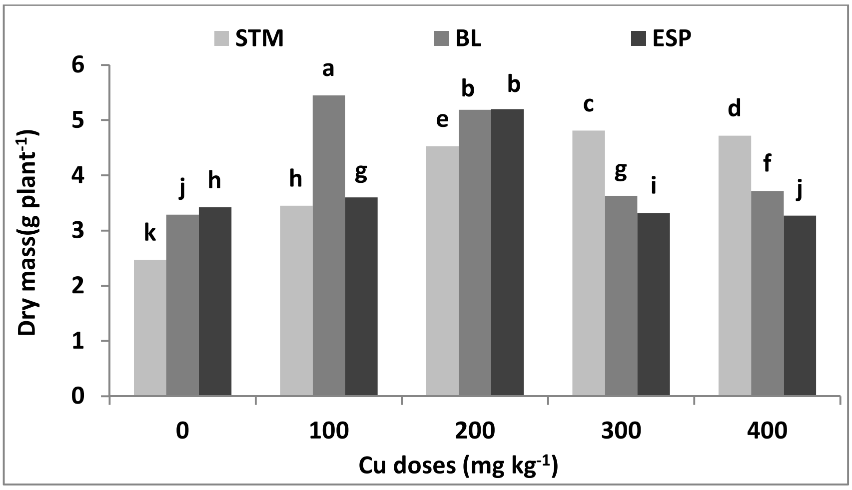

Fig. 5: Dry mass of culm of giant reed ecotypes (STM, BL, and ESP) at harvest after grown on elevated Cu concentrations. Different letters over columns showg significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Dry masses of leaf blade of STM, BL, and ESP ecotypes are presented in Fig. 6. Whilst, STM plants had lowest dry mass of leaf blade among all giant reed ecotypes, in higher Cu treatments above 200 mg kg^{-1} it had the highest dry mass compared to BL and ESP ecotypes. However, all ecotypes showed significant effects towards increasing Cu, where they measured higher values of leaf blade dry mass compared to control. In lower Cu concentrations than 300 mg kg^{-1}, BL and ESP plants had higher dry masses than STM plant, but BL ecotype was the highest ecotype in dry mass of leaf blade in Cu treatments up to 200 mg kg^{-1}.

3.2.2. Cu content in different plant parts of giant reed ecotypes

Total Cu content in shoot part of mother plant was measured (because separation of root system of mother plant from new tillers was impossible) and is depicted in Fig.

7. Significant differences ($p < 0.05$) of Cu contents of mother plant of all three studied ecotypes (STM, BL, and ESP) were found with increasing Cu in soil-like growth medium. Mother plant of STM ecotype had the highest content of Cu in its shoot parts compared to other two ecotypes under treatments of 100 and 300 mg Cu kg^{-1}, respectively (Fig. 7).

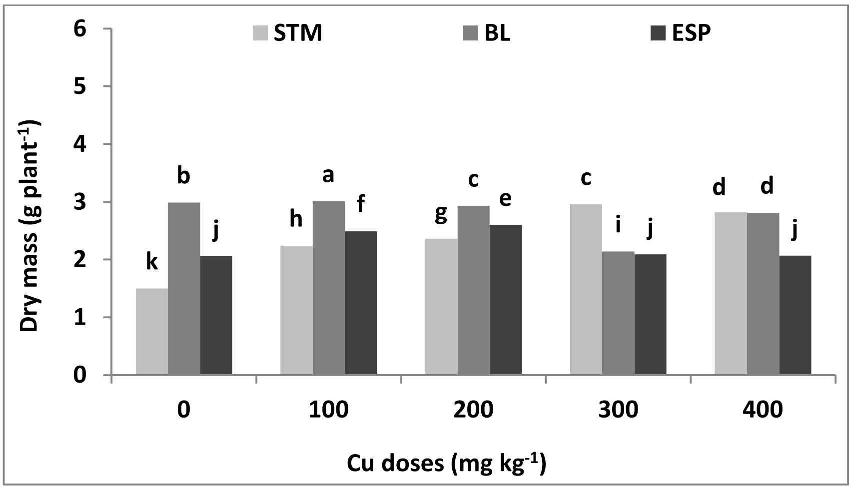

Fig. 6: Dry mass of leaf blade of giant reed ecotypes (STM, BL, and ESP) at harvest after grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

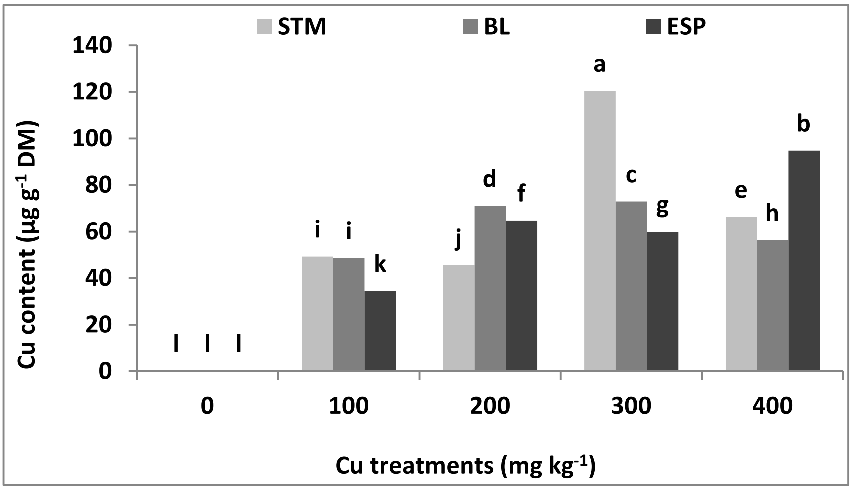

Fig. 7: Cu content of mother plant shoots of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Cu content in root system of investigated STM, BL, and ESP ecotypes of giant reed significantly increased with increasing Cu concentrations in experimental pots as shown in Fig. 8. However, all ecotypes accumulated increasing amounts of Cu within their root systems with increasing Cu level in treatments, where Cu content in root system at treatment of 400 mg Cu kg^{-1} was 3 folds more than Cu content in root system of studied ecotypes at treatment of 100 mg Cu kg^{-1}. Plants of ESP ecotype showed higher ability to uptake Cu from contaminated soil, where it had the highest content of Cu in root system compared to other ecotypes at treatments of 100, 300, and 400 mg kg^{-1}, achieving 549, 1631, and 1829 µg g^{-1} DM. In order to ensure complete cleaning for Cu contaminated soil, giant reed ecotypes have to be taken off with its root system from soil after phytoremediation (however, it is difficult and uneconomic to remove the total root system of giant reed planst, since it has extended network system of rhizomes) because the highest accumulated amount of Cu will be in root system. This also could explain why no foliar symptoms of Cu toxicity had been noticed on shoot system of giant reed ecotypes which used in this current study.

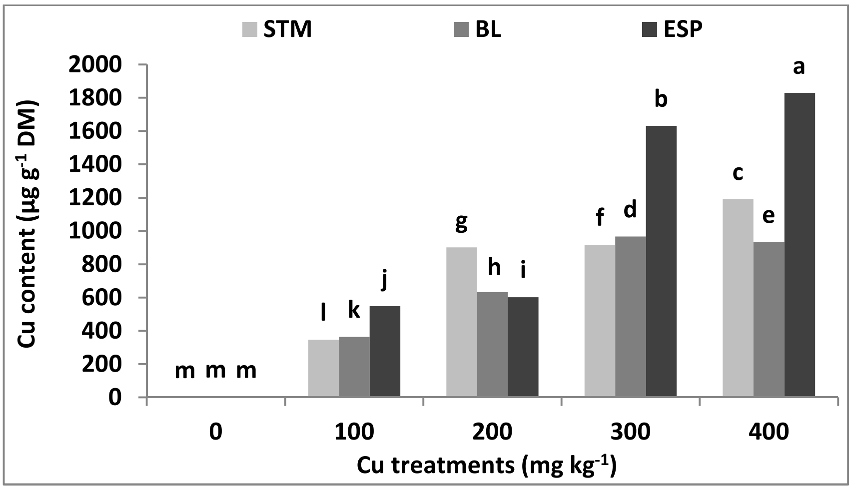

Fig. 8: Cu content of root system of giant reed ecotypes (STM, BL, and ESP) grown on different Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Cu content of culm of ESP plant was directly proportional to Cu doses in soil-like growth medium as seen in Fig. 9. As Cu increased in the treatments, culm of ESP plant concentrated more Cu in its tissues ranging from 43 to 101 µg g^{-1} DM at treatments of 100 to 400 mg Cu kg^{-1}, respectively. However, the highest content of Cu (130 µg g^{-1} DM) among all ecotypes was recorded for culm of BL ecotype at treatment of 200 mg kg^{-1}. STM plant had highest Cu content (113 µg g^{-1} DM) in its culm at treatment of 100 mg kg^{-1}. Generally, both STM and BL ecotypes did not show obvious trend towards increasing Cu in treatments, where hesitated tendency was noticed.

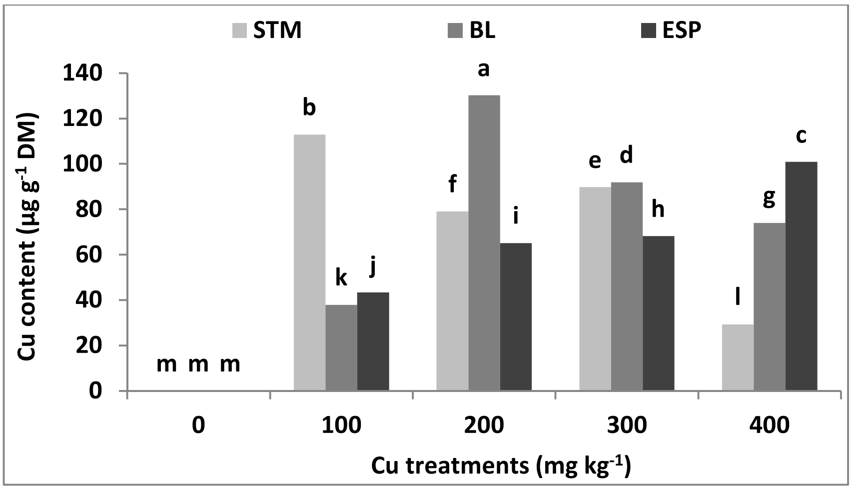

Fig. 9: Cu content of culm of giant reed ecotypes (STM, BL, and ESP) grown on different Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

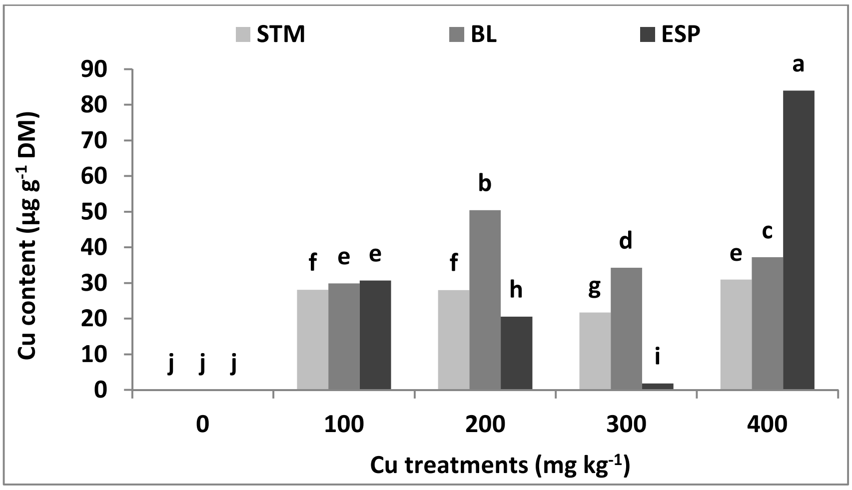

Fig. 10: Cu content of leaf blade of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Basically, increasing Cu levels did not substantially increase the contents of Cu in leaf blade of all investigated ecotypes, except ESP plant which had the highest recorded Cu content (84 µg g^{-1} DM) at highest Cu treatment, i.e., 400 mg kg^{-1}. However, at treatment of 300 mg Cu kg^{-1} leaf blade of ESP ecotype had the lowest measured value of Cu content (2 µg g^{-1} DM) among all ecotypes, but this could be due do mistake in sample preparation for measurements (Fig. 10). Broadly, significant differences were noticed between ecotypes within treatments, but no clear trend for increasing Cu on content of Cu in leaf blade. At treatments of 200 and 300 mg kg^{-1} BL ecotype recorded the highest Cu content in tissues of its leaf blade; likewise ESP ecotype had the highest Cu content at treatments of 100 and 400 mg kg^{-1}.

3.2.3. Cu uptake and phytoremediation indices

Basically, calculated values of transportation factor (TF) significantly affected with increasing Cu concentrations in soil-like growth medium. All TF values for STM, BL, and ESP ecotypes were less than one, which means that giant reed ecotypes poorly transported Cu from root system (underground part) to shoot system (aboveground part). This could point out that for complete and efficient phytoremediation process, removing the whole plant including its root system is mandatory. However, estimated TF values for all investigated ecotypes increased firstly then declined significantly with increasing Cu concentration above 200 mg kg^{-1} supposing transport channels of roots of giant reed plants were damaged above 200 mg Cu kg^{-1}.

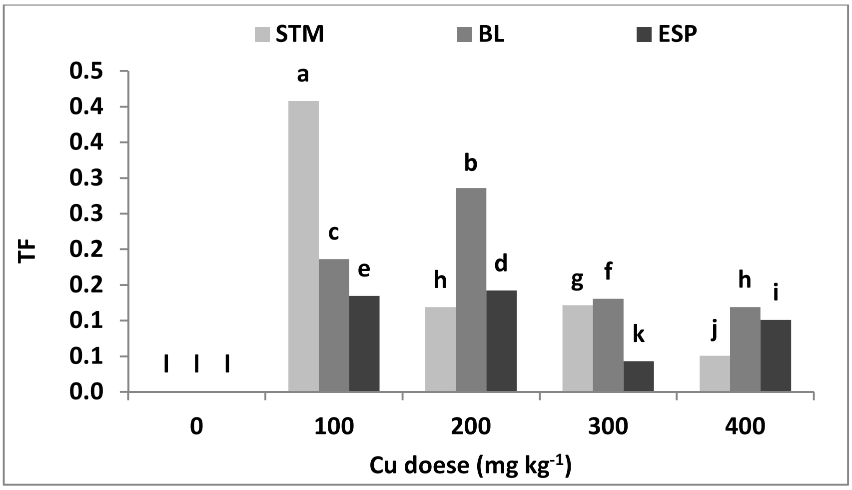

Fig. 11: Transportation factor (TF) for Cu uptake of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Conversely to TF values, estimated values of bioconcentration factor (BCF) were higher than one for all giant reed ecotype used in current study at all treatments of Cu. Increasing Cu concentration resulted in significant differences ($p < 0.05$) for BCF values among investigated ecotypes. The lowest measured BCF value was 2.6 recorded for BL plant when Cu concentration was 400 mg kg^{-1} (Fig. 12). However, calculated

BCF values for STM and BL ecotype firstly increased then significantly reduced with increasing Cu concentrations above 200 mg kg^{-1}. On the other hand, ESP plants were less affected negatively with increasing Cu levels in growth medium, where it recorded BCF values of 6.2, 3.4, 5.7 and 5.0 when Cu concentrations ranged from 100 to 400 mg kg^{-1}, respectively.

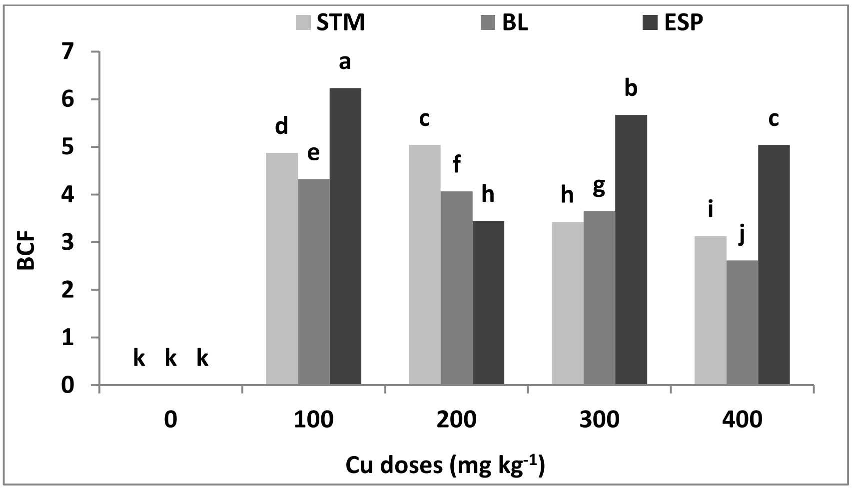

Fig. 12: Bioconcentration factor (BCF) for Cu uptake of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

Substantially, all studied ecotypes had same efficiency towards removing Cu from soil-like growth medium. Values of removal rate (RR) ranged from minimum 0.81 to maximum 0.89 regarding STM, BL, and ESP ecotypes (Fig. 13). However, with increasing Cu concentrations from 100 to 400 mg kg^{-1} no big differences were determined for RR under investigated giant reed ecotypes. Capacity of STM plant for removing declined when Cu increased in soil from 100 to 400 mg kg^{-1}, where RR values were 0.88, 0.82, 0.81, and 0.83, respectively. While, ESP plant possessed gradually reduced values for RR with increasing Cu levels in experimental pots, BL had no clear trend for its RR value where it had RR of 0.89 when Cu concentration was 100 mg kg^{-1} then decreased to reach 0.85 at treatment of 300 mg kg^{-1} but increased again to be 0.89 at highest Cu treatment (400 mg kg^{-1}). These results encourage the recommendation of giant reed ecotypes for Cu phytoremediation purposes, where they proved high efficiency and ability for theoretical removing Cu from Cu-contaminated site with some of 89%.

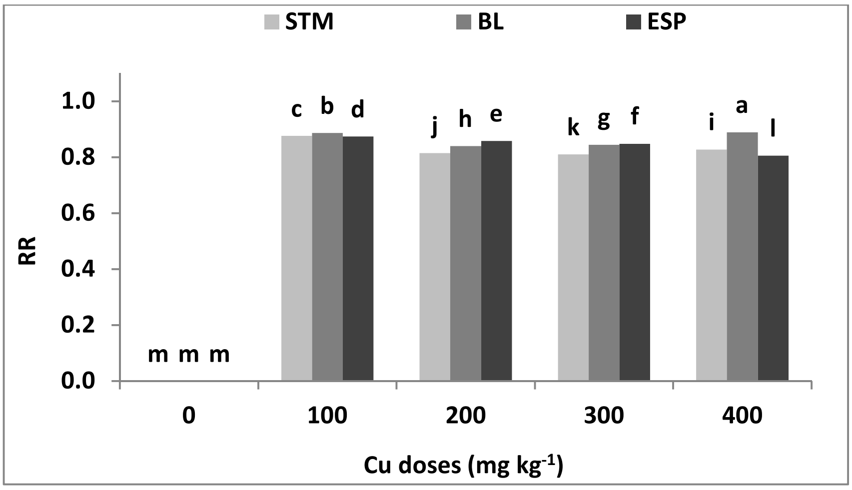

Fig. 13: Removal rate (RR) of Cu by giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over columns show significant differences among groups of treatments according to Duncan's test at $p < 0.05$.

3.2.4. Malondialdehyde content of giant reed ecotypes

MDA content of root systems of STM, BL, and ESP ecotypes significantly increased with adding Cu to soil-like growth medium compared to control plant. While, STM and ESP control plants had almost same content of MDA in their root systems, content of MDA of BL ecotype was nearly half of that found for STM and ESP ecotypes (Fig. 14). With applying Cu treatments, MDA content of root system in all ecotypes was closely same; this indicates that Cu treatments had stronger effect on root system of BL plant than STM and ESP plants. However, MDA content of STM root regularly increased with increasing Cu concentrations.Likewise, MDA content of root of ESP plant positively increased with elevating Cu levels. Response of BL ecotype for elevated Cu doses was slightly different from STM and ESP ecotypes. MDA content was 7.1 nmol g^{-1} DM for control plant and increased gradually recording 22.6 nmol g^{-1} DM when Cu concentration was 300 mg kg^{-1} then a small decreasing was measured when Cu concentration was 400 mg kg^{-1}.

MDA content in the culms of STM, BL, and ESP ecotypes grown under different concentrations of Cu was significantly enhanced versus the control (Fig. 15). At control treatment ESP culm had the highest MDA content compared to STM and BL, it measured almost 2 folds than STM and less than 2 folds for BL. However, when Cu doses increased in soil-like growth medium, MDA content of culm was gradually increased for all studied ecotypes.

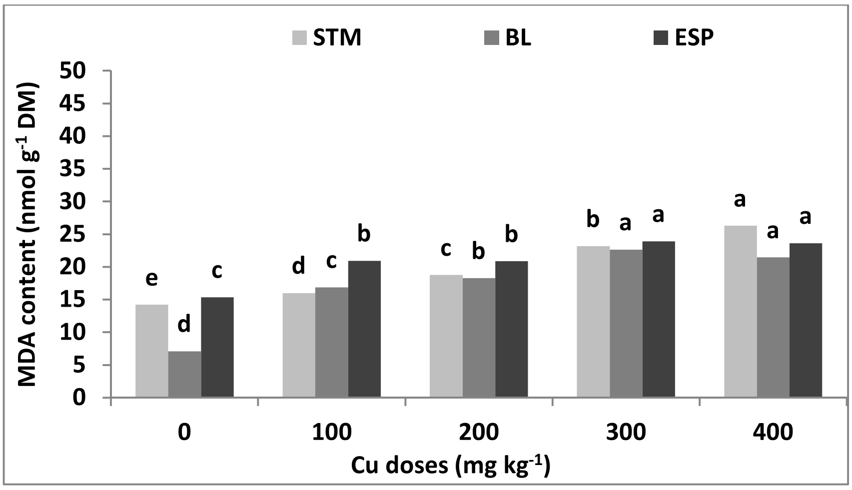

Fig. 14: Malondialdehyde (MDA) content in root system of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over same columns show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

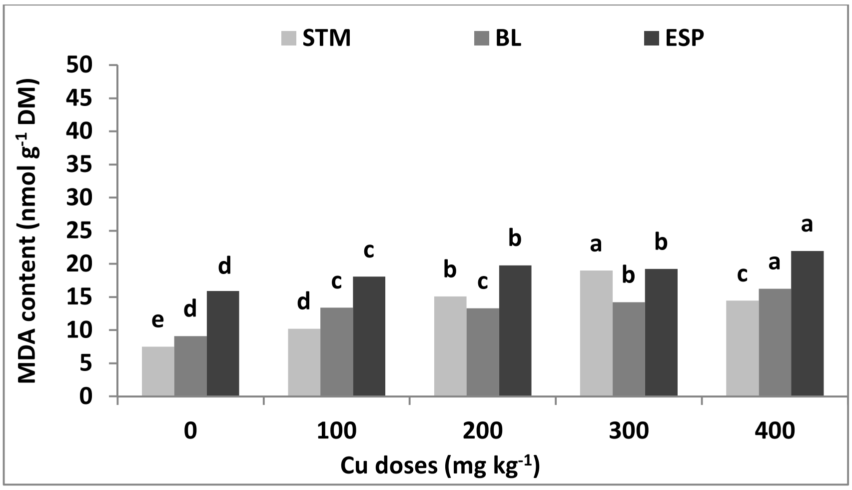

Fig. 15: Malondialdehyde (MDA) content in culm of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over same columns show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

Similarly, MDA content in the leaf blades of STM, BL, and ESP was significantly increased with increasing Cu levels. Leaf blade of control plants of all ecotypes had the lowest MDA content compared to Cu treatments. However, 9.8 nmol g^{-1} DM of MDA were measured in leaf blades of STM control plant but 19.5 nmol g^{-1} DM of MDA was found at the highest Cu treatment. On the other hand, leaf blades of BL had 11.9 and 18.2 nmol g^{-1} DM of MDA at treatments of 0 and 400 mg Cu kg^{-1}, respectively. Leaf blades of STM recorded the highest content of MDA at no Cu treatment achieving 18.7 nmol g^{-1} increased to 24.3 nmol g^{-1} DM when Cu was 400 mg kg^{-1} (Fig. 16).

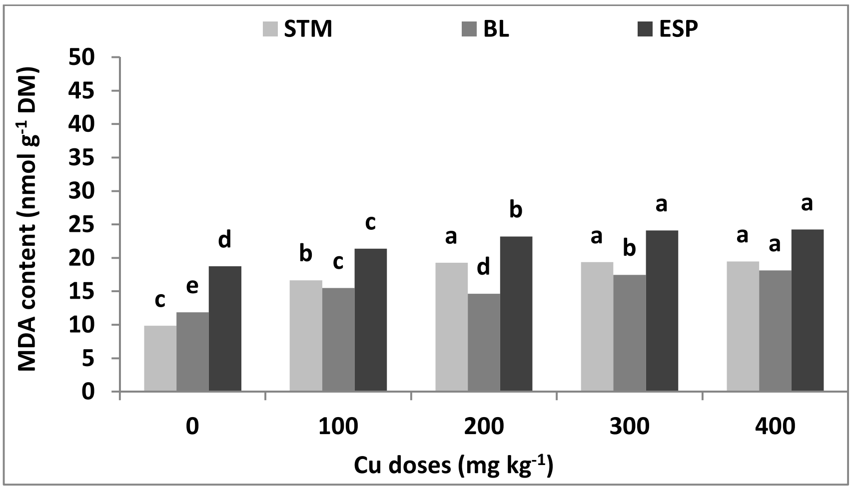

Fig. 16: Malondialdehyde (MDA) content in leaf blade of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over same columns show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

3.2.5. Chlorophylls *a* and *b* and carotenoids contents of giant reed ecotypes

Figure 17 displays chl *a* content in leaves of STM, BL, and ESP ecotypes. Clearly, increasing Cu doses had significant effect on chl *a* content in all ecotypes. Leaves of BL plant had the highest content of chl *a* at all Cu treatments compared to STM and ESP plants. However, chl *a* content in leaves of BL plant changed from 32 to 46 mg cm^{-2} when Cu concentration increased from 0 to 400 mg kg^{-1} in soil-like growth medium. Regular increase in chl *a* content was measured in leaves of STM plant when Cu concentration increased in soil-like growth medium up to 400 mg kg^{-1}. These findings illustrated that all giant reed ecotypes under investigation are tolerant to high Cu concentrations, therefore they could act as phytoremediation candidates for Cu-contaminated sites.

Chl *b* content in leaves of STM, BL, and ESP ecotypes was significantly increased with increasing Cu concentrations in soil-like growth medium. When Cu doses increased in soil-like growth medium up to 300 mg kg^{-1}, linear increase in chl *b* content in leaves of STM plant was measured. But an exponential increase was found for chl *b* content in leaves of STM when Cu concentration increased up to 400 mg kg^{-1} (Fig. 18). BL and ESP plants responded to elevated Cu concentrations positively.

Content of chl *b* in leaves of BL and ESP plants firstly increased with increasing Cu concentrations up to 300 mg kg^{-1}, and then slightly decreased with increasing Cu to 400 mg kg^{-1}.

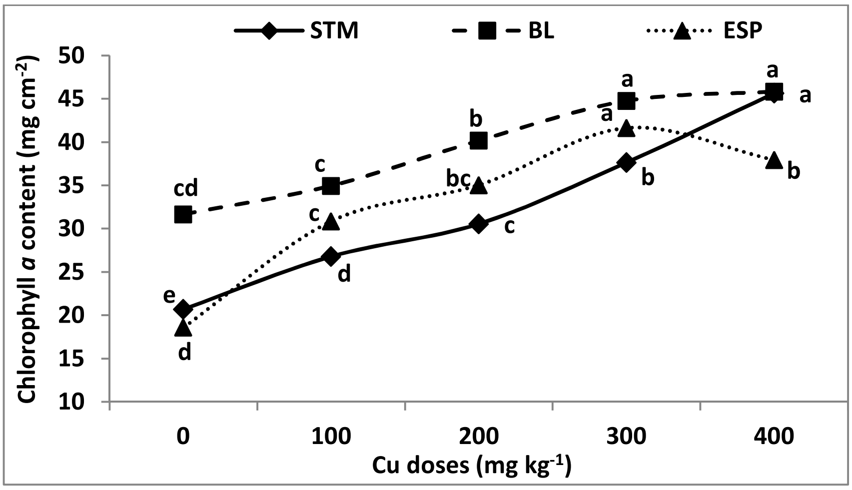

Fig. 17: Content of chlorophyll *a* in leaves of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over same columns show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

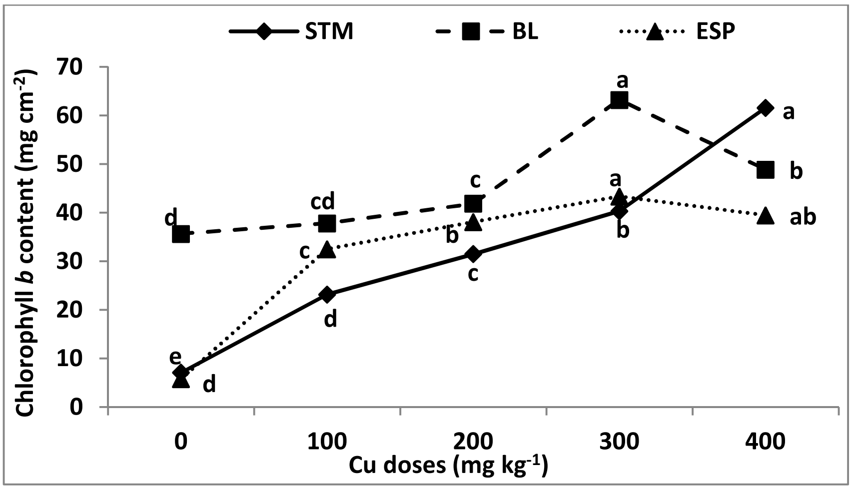

Fig. 18: Content of chlorophyll *b* in leaves of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over same columns show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

Carotenoids contents (µg cm^{-2}) in leaves of growing giant reed ecotypes on elevated Cu concentrations are depicted in Fig. 19. Positively increasing Cu levels enhanced carotenoid contents in leaves of studied giant reed ecotype. Significant differences were measured for carotenoid contents with elevating Cu doses. Content of carotenoid in leaves of both BL and ESP plants steadily induced with increasing Cu concentrations up to 300 mg kg^{-1} then decreased when Cu concentration was 400 mg kg^{-1}. In leaves of STM plant carotenoid content linearly increased with increasing Cu concentrations, where highest content of carotenoid was 19 µg cm^{-2} when Cu concentration was 400 mg kg^{-1}. However, treatments of 100 and 200 mg Cu kg^{-1} did not resulted in siginificant differences of carotenoid content.

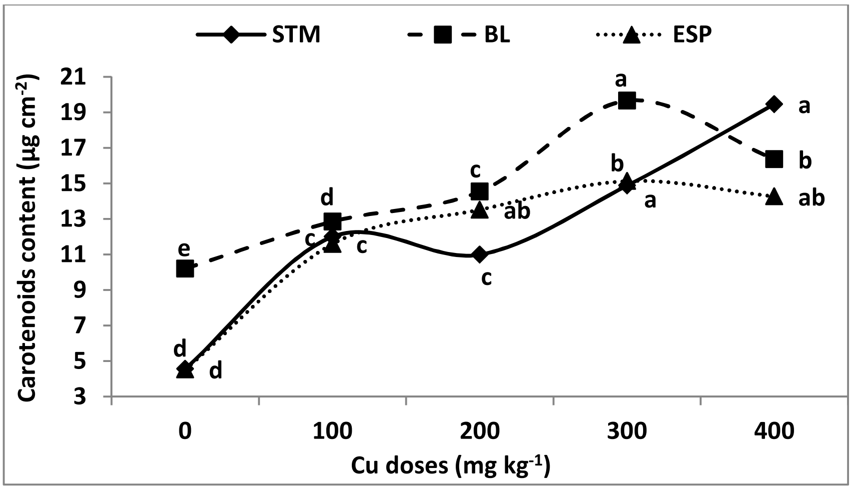

Fig. 19: Content of carotenoids in leaves of giant reed ecotypes (STM, BL, and ESP) grown on elevated Cu concentrations. Different letters over same columns show significant differences among each group of treatments according to Duncan's test at $p < 0.05$.

3.2.6. Efficiency of photosynthesis under elevated Cu exposures

At the physiological level, the measurement of photosynthetic efficiency (Fv/Fm) is useful and effective parameters to assess the photosynthetic status of plants under heavy metals stress conditions. Table 2 showed values of Fv/Fm measured in leaves of STM, BL, and ESP ecotypes after growing on elevated Cu concentrations. Greenhouse experiment has been started in mid of July 2013 and one and half months later (September) Fv/Fm was measured and after one month (October) it was measured again. However, with respect to control plants no significant differences were measured at Cu treatments for all investigated ecotypes of giant reed. In September, the highest values of Fv/Fm of STM, BL, and ESP ecotypes were found at the highest Cu treatment (400 mg kg^{-1}). Values of Fv/Fm of STM plant ranged from 0.80 to 0.83 at treatments of 0 to 400 mg Cu kg^{-1}. While, BL plants had values changed from 0.81 to 0.84, ESP plants recorded values of 0.81 to 0.81 when Cu concentrations changed from 0 to 400 mg kg^{-1}. In general, values of Fv/Fm that measured in October were higher than that measured in September within all ecotypes, but this increase was not significant. As a consequence for non-siginificant values of Fv/Fm that measured under different Cu treatments with respect to control, it could be concluded that giant reed ecotypes did not negatively affected with high Cu concentrations, but have been enhanced therefore they showed tolerant characteristics for elevated Cu levels.

Table 2: Maximum quantum efficiency of PSII (Fv/Fm) of dark adapted leaves of giant reed ecotypes treated with elevated Cu concentrations.

Cu doses	September			October		
(mg kg^{-1})	*STM*	*BL*	*ESP*	*STM*	*BL*	*ESP*
0	0.80cde	0.81abcd	0.81bcde	0.83ab	0.83ab	0.83ab
100	0.83ab	0.83a	0.80de	0.82abc	0.83ab	0.79d
200	0.82abcd	0.82abcd	0.81bcde	0.83ab	0.83ab	0.81bcd
300	0.83ab	0.82abcd	0.79e	0.83ab	0.84a	0.80cd
400	0.83abc	0.84a	0.81bcde	0.83a	0.84a	0.82ab

In columns different letters showing significant differences among each group of treatments according to Duncan's test at $p < 0.05$

4. CONCLUSIONS AND RECOMMENDATIONS

4.1. *In vitro* experiment

- Giant reed ecotypes (BL and 20SZ) which are used in the present study showed great tolerance to elevated Cu concentrations up to 26.8 mg L^{-1} in artificially Cu-contaminated water.

- No Cu toxicity symptoms were reported on shoot and root parts of both ecotypes up to 10 mg Cu L^{-1}, but some reduction in root systems were noticed at treatment of 26.8 mg Cu L^{-1}.
- Both giant reed ecotypes showed capacity to uptake Cu from artificially contaminated water bodies, where the removal rates ranged between 96.6 to 100%.
- Almost 50% from total uptake Cu content was found in root system of both ecotypes.

- Transportation and bioconcentration factors had values higher than 1 especially whec Cu concentration was higher than 5 mg L^{-1}.
- Both ecotypes showed considerable and significant efficiency to clean Cu-contaminated water bodies up to 26.8 mg L^{-1}. So, it is a positive indicator of their potential capacity to serve as a phytoremediation plant together with the existence of nontoxic symptoms on the plants. These results emphasized that both ecotypes being nonfood crop, might act as good phytoremediation candidates for Cu-contaminated water bodies.

4.2. *Ex vitro* experiment

- STM, BL, and ESP ecotypes of giant reed which derived by somatic embryogenesis were normally grown on elevated Cu concentrations up to 400 mg kg^{-1}.
- No foliar symptoms for Cu toxicity were seen on aboveground part (shoot) of any ecotype.
- Root system of all ecotypes was significantly reduced at the highest Cu concentration (400 mg kg^{-1}).
- Shoot length of all ecotypes was significantly enhanced with increasing Cu levels in soil-like growth medium.
- Dry mass of root system of giant reed ecotypes was slightly decreased with inceasing Cu doses with respect to control plant, while dry mass of culm and leaf blade was significantly enhanced when Cu concentrations increased in soil-like growth medium. All three ecotypes showed same efficiencies for uptake Cu, where they removed 82.8 to 88.0 % from total Cu under different Cu treatments.
- Most of removed Cu was accumulated in root system of giant reed ecotypes.
- At higher Cu concentrations above 100 mg kg^{-1}, BL plant had higher transportation factor (TF) than STM and ESP, but all values of TF were less than 1.
- Bioconcentration factor (BCF) values were higher than reference value of 1, and ESP recorded the higher values with respect to STM and BL plants.
- Photosynthesis pigments such as chlorophylls *a* and *b* as well as carotenoids content were significantly enchanced with increasing Cu levels.

- Total protein contents in different plant parts of all ecotypes were induced significantly when Cu levels increased in soil-like growth medium.
- Increasing Cu concentrations negatively affected malondialdehyde (MDA) content in all plant parts of STM, BL, and ESP ecotypes, where higher MDA contents were measured under Cu treatments versus control plants.

From all the metioned data above, it could be concluded that giant reed ecotypes showed considerable ability and capacity to grow on elevated Cu concentrations. Also they have great potential for removing Cu from Cu-contaminated soil-like growth medium. Therefore, it could be recommended that giant reed ecotypes can be employed as phytoremediation candidate as well as producing significane biomass production during remediation process.

5. NEW SCIENTIFIC RESULTS

- Using giant reed ecotypes for decontaminating Cu-contaminated water bodies due to its ability to translocate the Cu from roots to shoots.
- Industerial wastewater polluted with Cu can be restored by giant reed ecotypes, where they showed significant ability to remove Cu from wastewater reached some of 92 to 99%.
- In soil-like environemnts which are contaminated with high Cu levels up to 400 mg kg^{-1}, giant reed ecotypes can remediate it with efficiencies ranged between 82.6 to 88.0%.
- Photosynthesis pigments positively enhanced by elevated Cu concentrations; this indicates that studied giant reed ecotypes are tolerant to high Cu levels up to 400 mg kg^{-1}.
- High Cu exposures did not negatively affect the biomass production of giant reed plants; therefore considerable biomass production is expected for giant reed ecotypes during the phytoremediation process as an additional benefit.

6. SCIENTIFIC RESULTS UTILIZABLE IN THE PRACTICE

- Giant reed plants showed considerable effects to remediate the Cu-contaminated streams
- The possibility of using giant reed plants in Cu-contaminated soil-like growth medium, since it showed high tolerance for growing on increasing Cu levels.
- Giant reed is considered as good candiate for restoring and remediating Cu-contaminated environments, where it showed great capacity and efficiency for uptake Cu from contaminated soil-like growth medium.
- Using giant reed as bioenergy crop during the phytoremediation process, where it can produce significant biomass production especially on Cu-contaminated sites.

7. REFERENCES

Márton L. – Czakó M. (2002): Szelektált egyszikü nemzetségek hosszan tartó totipotens tenyészete. [Magyar szabadalmi bejelentés P0500786]

Márton L. – Czakó M. (2004): Sustained Totipotent Culture of Selected Monocot Genera. USA, patent 6,821,782.

Márton L. – Czakó M. (2007): Sustained Totipotent Culture of Selected Monocot Genera. USA, patent 7,303,916.

Wu Qi Z. S. – Cheng L. – Wu G. (2012): Physiology and phytoaccumulation of *Arundo donax* subjected to exogenous copper. Acta Laser Biol Sintica. **21**. 4. 360–364. doi:10.3969 /j.issn.1007-7146.2012.04.013

8. PUBLICATIONS

~~Registry number: DEENK/89/2015.PL~~
Subject: Ph.D. List of Publications

Candidate: Nevien Adel Elhawat
Neptun ID: Y50KNA
Doctoral School: Kerpely Kálmán Doctoral School of Corp Production, Horticulture and Regional Sciences

List of publications related to the dissertation

Foreign language international book chapter(s) (4)

1. Alshaal, T., **Elhawat, N.**, Domokos-Szabolcsy, É., Kátai, J., Márton, L., Czakó, M., El-Ramady, H., Fári, M.G.: Giant Reed (Arundo donax L.): A Green Technology for Clean Environment.
   In: Phytoremediation: Management of Environmental Contaminants. Ed.: A.A. Ansari et al., Springer Science + Business Media, LLC, Switzerland, 3 - 20, 2015. ISBN: 9783319103945
   DOI: http://dx.doi.org/10.1007/978-3-319-10395-2_1

2. El-Ramady, H.R., Alshaal, T.A., Shehata, S.A., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Fári, M., Márton, L.: Plant Nutrition:From Liquid Medium to Micro-farm.
   In: Sustainable Agriculture Reviews volume 14: Agroecology and Global Change. Ed.: Harry Ozier-Lafontaine, Magalie Lesueur-Jannoyer, Springer, Cham; Heidelberg; New York; Dordrecht; London, 449-508, 2014. ISBN: 9783319060156
   DOI: http://dx.doi.org/10.1007/978-3-319-06016-3

3. El-Ramady, H.R., Alshaal, T.A., Amer, M., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Fári, M.: Soil Quality and Plant Nutrition.
   In: Sustainable agriculture aeviews volume 14: Agroecology and Global Change. Ed.: Harry Ozier-Lafontaine, Magalie Lesueur-Jannoyer, Springer, Cham; Heidelberg; New York; Dordrecht; London, 345-448, 2014. ISBN: 9783319060156
   DOI: http://dx.doi.org/10.1007/978-3-319-06016-3

4. Alshaal, T.A., El-Khir, A.M.A., **Elhawat, N.**, El-Essawi, T.M., El-Ramady, H.R.: Integrated nutrient management and soil quality.
   In: Contemporary environmental readings: towards sustainability for agriculture. Hassan R. El-Ramady, Naama A. Abd Alla, Said A. Shehata, LAP LAMBERT Academic Publishing, Germany, 1-121, 2013. ISBN: 9783659342905

Address: 1 Egyetem tér, Debrecen 4032, Hungary Postal address: Pf. 39. Debrecen 4010, Hungary
Tel.: +36 52 410 443 Fax: +36 52 512 900/63847 E-mail: publikaciok@lib.unideb.hu, ¤ Web: www.lib.unideb.hu

Foreign language scientific article(s) in Hungarian journal(s) (1)

5. **Elhawat, N.**, Alshaal, T., Kratz, S., Domokos-Szabolcsy, É., El-Ramady, H., Prokisch, J., Eszenyi, P., Sztrik, A., Babka, B., Fári, M.: Ecotoxicology of Copper in Horticultural Soils: A Review.
Int. J. Hortic. Sci. 19 (1-2), 7-18, 2013. ISSN: 1585-0404.

Foreign language scientific article(s) in international journal(s) (3)

6. **Elhawat, N.**, Alshaal, T., Domokos-Szabolcsy, É., El-Ramady, H., Márton, L., Czakó, M., Kátai, J., Balogh, P., Sztrik, A., Molnár, M., Popp, J., Fári, M.G.: Phytoaccumulation potentials of two biotechnologically propagated ecotypes of Arundo donax in copper-contaminated synthetic wastewater.
Environ. Sci. Pollut. Res. 21 (12), 7773-7780, 2014. ISSN: 0944-1344.
DOI: http://dx.doi.org/10.1007/s11356-014-2736-8
IF:2.757 (2013)

7. Alshaal, T., Domokos-Szabolcsy, É., Márton, L., Czakó, M., Kátai, J., Balogh, P., **Elhawat, N.**, El-Ramady, H., Gerőcs, A., Fári, M.: Restoring Soil Ecosystems and Biomass Production of Arundo donax L. under Microbial Communities-Depleted Soil.
Bioenerg. Res. 7 (1), 268-278, 2013. ISSN: 1939-1234.
DOI: http://dx.doi.org/10.1007/s12155-013-9369-5
IF:3.398

8. Alshaal, T., Domokos-Szabolcsy, É., Márton, L., Czakó, M., Kátai, J., Balogh, P., **Elhawat, N.**, El-Ramady, H., Fári, M.: Phytoremediation of bauxite-derived red mud by giant reed.
Environ. Chem. Lett. 11 (3), 295-302, 2013. ISSN: 1610-3653.
DOI: http://dx.doi.org/10.1007/s10311-013-0406-6
IF:1.906

Hungarian conference proceeding(s) (3)

9. **Elhawat N.**, Alshaal T., Domokos-Szabolcsy É., El-Ramady H., Antal G., Márton L., Czakó M., Balogh P., Fári M.: Biomassza termelés alternatív lehetősége rézzel szennyezett talajokon. In: XXI. Növénynemesítési Tudományos Napok : összefoglalók. Kiadta: a Magyar Tudományos Akadémia, Magyar Tudományos Akadémia, Martonvásár, 109, 2015.

Address: 1 Egyetem tér, Debrecen 4032, Hungary Postal address: Pf. 39. Debrecen 4010, Hungary
Tel.: +36 52 410 443 Fax: +36 52 512 900/63847 E-mail: publikaciok@lib.unideb.hu, ¤ Web: www.lib.unideb.hu

10. Alshaal T., Domokos-Szabolcsy É., Márton L., Kátai J., **Elhawat N.**, Antal G., Gerőcsi A., Fári M.: Az olasznád (Arundo donax L.) szomatikus növények talaj mikrobiális közösségei és szerepük a fitoremediációs folyamatokban.
In: The proceeding of XIX. Növénynemesítési Tudományos Nap. Szerk.: Hoffmann Borbála, Kollaricsné Horváth Margit, Pannon Egyetem Georgikon Kar, Keszthely, 72, 2013.

11. **Elhawat N.**, Domokos-Szabolcsy É., Alshaal T., Molnár M., Antal G., Márton L., Fári M.: Szomatikus embrió eredetű olasznád (Arundo donax L.) klaszterek in vitro sótűrése két ökotípus összehasonlításával.
In: The proceeding of the XIX. Növénynemesítési Tudományos Nap : összefoglalók. Szerk.: Hoffmann Borbála, Kollaricsné Horváth Margit, Pannon Egyetem Georgikon Kar, Keszthely, 85, 2013. ISBN: 9789639639508

Foreign language conference proceeding(s) (9)

12. Alshaal, T., **Elhawat, N.**, El-Ramady, H., Domokos-Szabolcsy, É., Kátai, J., Márton, L., Czakó, M., Fári, M.: Influence of Giant Reed (Arundo donax L.) Plantation on Soil Quality in Different Regions: A Comparison Study.
In: Proceedings of the 11th International Conference "Climate Changes and Sustainable Development of Natural Resources. Ed.: by IUSS, International Union of Soil Sciences, Kafrelsheikh, Egypt, 80, 2014].

13. El-Ramady, H., Abd Alla, N., Alshaal, T., **Elhawat, N.**, Domokos-Szabolcsy, É., Prokisch, J., Sztrik, A., Fári, M.: Selenium: Its Implications to Human Health through Soils under Climate Changes.
In: Proceedings of the 11th International Conference "Climate Changes and Sustainable Development of Natural Resources. Ed.: by IUSS, International Union of Soil Sciences, Kafrelsheikh, Egypt, 54, 2014.

14. Domokos-Szabolcsy, É., El-Ramady, H.R., Alshaal, T., Márton, L., Zsiros, O., Garab, G., Veres, S., Prokisch, J., **Elhawat, N.**, Abd Alla, N., Fári, M.: Tolerance Efficiency and Soil Remediation Potential of Giant Reed (Arundo donax L.).
In: Proceedings of the 11th International Conference "Climate Changes and Sustainable Development of Natural Resources. Ed.: by IUSS, International Union of Soil Sciences, Kafrelsheikh, Egypt, 55, 2014.

15. El-Ramady, H.R., Abd Alla, N.A., Alshaal, T.A., **Elhawat, N.**, Domokos-Szabolcsy, É., Prokisch, J.: Nano-selenium: From in vitro to micro farm experiments.
In: Proceedings of the International Conference on Biogeochemical processes at air-soil-water interfaces and environmental protection. Ed.: by ESSC, European Society for Soil Conservation, [Imola-Ravenna, Italy], 76, 2014.

16. Kurucz, E., Domokos-Szabolcsy, É., Antal, G., Kaprinyák, T., Alshaal, T., **Elhwat, N.**, Abd Alla, N., El-Ramady, H., Fári, M.: Biotechnology assisted breeding of endangered virginia mallow (Sida hermaphrodita L.) in Central Europe.
In: International Conference : Climate changes and sustainable development of natural resources : book of abstracts. Ed.: by Kafrelsheikh University, Kafrelsheikh University, Egypt, 81-82, 2014.

17. Fári, M., Antal, G., Kurucz, E., Kaprinyák, T., Alshaal, T., **Elhwat, N.**, Abd Alla, N., El-Ramady, H., Domokos-Szabolcsy, É.: Biotechnology of new dedicated biomass crops: Plantbiogen program in Hungary.
In: International Conference : Climate changes and sustainable development of natural resources : book of abstracts. Ed.: by Kafrelsheikh University, Kafrelsheikh University, Egypt, 22, 2014.

18. Fári, M.G., Antal, G., Alshaal, T., **Elhawat, N.**, El Ramady, H., Kurucz, E., Domokos-Szabolcsy, É.: Biotechnology of Biomass Supply Chain:Central European Perspective.
In: Proceedings of the Pannonian Plant Biotech Association Workshop "Plant for the future". Ed.: Pannonian Plant Biotech Association, 14-15,

19. **Elhawat, N.**, Alshaal, T., Domokos-Szabolcsy, É., Kátai, J., Márton, L., Czakó, M., Molnár, M., Fári, M.G.: Comparative performance of copper uptake by two ecotypes of giant reed (Arundo donax L.) grown on hydroponic culture.
In: Proceedings of the Pannonian Plant Biotech Association Workshop "Plant for the future". Ed.: by Pannonian Plant Biotech Association, Pannonian Plant Biotech Association, [Cluj-Napoca], 33-34, 2013.

20. Alshaal, T., **Elhawat, N.**, Domokos-Szabolcsy, É., Kátai, J., Márton, L., Czakó, M., Gerőcs, A., Fári, M.G.: Growth dynamics and biomass production of Giant Reed under different watering regimes.
In: Proceedings of the Pannonian Plant Biotech Association Workshop "Plant for the future". Ed.: by Pannonian Plant Biotech Association, Pannonian Plant Biotech Association, [Cluj-Napoca], 31-32, 2013.

List of other publications

Foreign language scientific article(s) in Hungarian journal(s) (4)

21. Shehata, S.A., Hashem, M.Y., Mahmoud, G.I., Abd El-Gawad, K.F., El-Ramady, H., Alshaal, T., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Fári, M.: Effects of Foliar Nutrition on Onion Seed Storage under Controlled Atmosphere.
Int. J. Hortic. Sci. 19 (1-2), 101-110, 2013. ISSN: 1585-0404.

22. Shehata, S.A., Hashem, M.Y., Mahmoud, G.I., Abd El-Gawad, K.F., El-Ramady, H., Alshaal, T., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Fári, M.: Effect of Foliar Nutrition on Post-Harvest of Onion Seed under Sandy Soil and Saline Irrigation Water Conditions.
Int. J. Hortic. Sci. 19 (1-2), 85-92, 2013. ISSN: 1585-0404.

23. Shehata, S.A., Hashem, M.Y., Mahmoud, G.I., Abd El-Gawad, K.F., El-Ramady, H., Alshaal, T., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Fári, M.: Effects of Foliar Nutrition on Onion Seed Storage under Modified Atmosphere Packages.
Int. J. Hortic. Sci. 10 (1-2), 93-100, 2013. ISSN: 1585-0404.

24. Shehata, S.A., Hashem, M.Y., Mahmoud, G.I., Abd El-Gawad, K.F., El-Ramady, H., Alshaal, T., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Fári, M.: Foliar Nutrition and Post-Harvest of Onion Seed: Effects of storage temperatures, storage period and foliar nutrition.
Int. J. Hortic. Sci. 19 (1-2), 29-47, 2013. ISSN: 1585-0404.

Foreign language scientific article(s) in international journal(s) (1)

25. El-Ramady, H., Abdalla, N., Alshaal, T., Domokos-Szabolcsy, É., **Elhawat, N.**, Prokisch, J., Sztrik, A., Fári, M., El-Marsafawy, S., Shams, M.S.: Selenium in soils under climate change, implication for human health.
Environ. Chem. Lett. 13 (1), 1-19, 2014. ISSN: 1610-3653.
DOI: http://dx.doi.org/10.1007/s10311-014-0480-4
IF:1.906 (2013)

Foreign language conference proceeding(s) (2)

26. Fári, M., Antal, G., Kurucz, E., Kaprinyák, T., Alshaal, T., **Elhwat, N.**, Abd Alla, N., El-Ramady, H., Domokos-Szabolcsy, É.: Bioipari célra nemesített évelő biomassza növények kutatása Debrecenben: Plantbiogen program = Research on dedicated perennial biomass crops in Debrecen : the plantbiogen program.
In: XX. Növénynemesítési Tudományos Nap : Növénynemesítés a megújuló mezőgazdaságban. Szerk.: Veisz Ottó, A Magyar Tudományos Akadémia Agrártudományok Osztályának Növénynemesítési Tudományos Bizottsága, [Budapest], 140-144, 2014. ISBN: 9789638351425

27. El-Ramady, H.R., Alshaal, T., Amer, M., Aiad, M., **Elhawat, N.**: Significance of nutrient and water sustainability: Effect of land leveling, cut off irrigation and N- fertilization on maize yield.
In: Proceedings of International Conference of German Soil Science Society (BDG). BDG, Göttingen, 1-4, 2013.

Total IF of journals (all publications): 9,967
Total IF of journals (publications related to the dissertation): 8,061

The Candidate's publication data submitted to the iDEa Tudóstér have been validated by DEENK on the basis of Web of Science, Scopus and Journal Citation Report (Impact Factor) databases.

23 April, 2015