IJFANS INTERNATIONAL JOURNAL OF FOOD AND NUTRITIONAL SCIENCES
ISSN PRINT 2319 1775 Online 2320 7876
 UGC CARE Listed ( Group -I) Journal Volume 12, Iss 1, 2023

# Tolerance of nickel by Indian mustard genotypes from Ni enriched soil

**Sonal Vishnoi [1*], Manju Bala [2], and Anu Godara [3]**

**[1] Associate Professor, Department of Chemistry, Govt. P.G. Collage, Hisar (Haryana) Mobile: 9466760205**
**Email.id: sonalvishnoi.sv.@gmail.com**

**[2] Guest Lecturers, Department of Applied Chemistry, Govt. Polytechnic Sonipat (Haryana) Mobile: 8950099999**
**Email.id: anilbhaddu@gmail.com**

**[3]Research Scholar, Department of Chemistry, JCDV Memorial college, Sirsa (Haryana) Mobile: 9588307116**
**Email.id: ankurbishnoi729@gmail.com**

***Corresponding author: Email.id: sonalvishnoi.sv.@gmail.com Mb. 9466760205**

## ABSTRACT

A pot experiment was conducted to investigate the tolerance of nickel by three Indian mustard plant genotypes, namely, RH - 819, Varuna and Rh-9304, in a light soil from sand dune areas of Balsamand, Hisar. Five levels of Ni concentration ranging from 0-120 mg $kg^{-1}$ soil were taken for the study. The toxicity symptoms were recorded; biomass production, Ni concentration and finally the Ni uptake were measured to screen the best Ni tolerant Indian mustard genotype. The plants were harvested at maturity. The results of this study showed that all the three Indian mustard genotypes survived upto Ni 200 mg $kg^{-1}$ soil level. The genotypes exhibited visual Ni toxicity symptoms with narrow and small leaves, stunted growth, poor branching, delayed flowering and poor seed development and resulted into low biomass yield. Genotype RH-819 produced highest mean biomass, Ni concentration and its uptake on Ni spiked soil. Hence, genotype RH-819 of Indian mustard was found the best Cd and Ni tolerant genotype and was selected for uptake study.

***Key words:*** **Chelator, Tolerance, Nickel uptake, Chlorophyll content, Heavy metal, toxicity, Indian mustard**

## INTRODUCTION

Over the course of recent decades, urbanization, industrial and agricultural activities have led to a continuous production of huge amount of heavy metals contaminated solid, liquid and as fine particles directly into atmosphere and ultimately deposited on the surface of land and water bodies which finally on reaching the agricultural fields get accumulated in soil at hazardous levels (Al-Hawari and Mulligan, 2006). The heavy metals contaminated soils are often used to grow vegetables i.e. leafy and tuber crops

 

and fodder to meet the demands of nearby urban areas. These crops are notorious for their capacity to accumulate heavy metals in their edible parts. In this way, the transfer of heavy metals to man may result from consuming contaminated edible parts of the plants or indirectly from consuming milk or meat from grazing animals that have consumed contaminated plants and ultimately the heavy metals accumulating in soil may get entry into the human and animal food chain and affecting plants, animals and human health (Bishnoi *et al.*, 1993b and Ahmed *et al.*, 2001). Thus, the issue of contamination of agricultural soils and chemical residues in agricultural produce have become increasingly important in recent years due to increased public awareness and concern for food and land quality (Okoronkwo *et.al.*, 2005).

In Haryana also large amount of sewage water and industrial effluent is produced every day which is used as a potential source of irrigation of fields. Long term application of effluent for irrigating crops may cause potentially toxic metal accumulation in soil to such an extent that they may cause toxic effect to plant growth. Soil contamination by heavy metals is of major concern because of their toxicity and threat to both human health and environment.

Hence, there is a need to develop suitable biological soil remediation technique to remove contaminants. In fact, traditional state-of-the-art technology for the remediation of metal polluted soils is the excavation and burial of the soil at a hazardous waste site. However, these approaches are expensive, disruptive, and are not economically viable. Recently, efforts have been made towards finding remediation strategies that are less expensive and less damaging to soil properties than current approaches. One such method is phytoextraction in which plants uptake heavy metal from the soil, followed by harvesting the above ground biomass. Harvested material is disposed in brick kilns (as bio-energy source) and byproduct in a landfill (kilns ash) or also treated to recover metals (Coopper *et al.*, l999). Use of chelating agents to enhance heavy metal uptake is another new line in the technique of phytoremediation. To be successful on a specific site, the remediation technique must be selected according to heavy metals on the soil particles. Some scientists recommended the use of hyperaccumulator species, other prefer plants with a lower accumulation rate but high biomass. Amongst the commercial crops grown in Haryana rabi season, Indian mustard has been reported to produce high biomass.

## MATERIALS AND METHODS

A pot experiment was conducted to investigate the Ni phytoextraction potential of three Indian mustard plant genotypes, namely, RH - 819, Varuna and Rh-9304, in a light soil from sand dune areas of Balsamand, Hisar. Five levels of NI concentration ranging from 0-120 mg $kg^{-1}$ soil were taken for the study. The toxicity symptoms were recorded; biomass production, Ni concentration and finally the Ni

 

uptake were measured to screen the best Ni tolerant Indian mustard genotype. The plants were harvested at maturity. The chlorophyll (a & b and total content) was extracted as per standard procedure of Hiscox and Israestam (1979). Eighty mg of washed and fine chopped leaf tissue was placed in a test tube containing 7 ml of DMSO (Di Methyl Sulphoxide). The chlorophyll was extracted without grinding by incubating at $65^0$C for 1 hour. The extracted liquid was transferred to a graduated cylinder and volume made upto 10 ml with DMSO and optical density was recorded using spectrophotometer at 645 and 663 nm. Chlorophyll content was calculated following the standard equation proposed by Arnon (1949) as follows: Chl 'a' (mg/g) = 11.63 x $A_{663}$ –2.39 x $A_{645,}$ Chl 'b' (mg/g) = 20.11x $A_{645}$ –5.18 x $A_{663,}$ Total Chlorophyll (mg/g) = 6.45 x $A_{663}$ +17.72 x $A_{645}$

**Estimation of nickel:** In order to determine Ni in plant bio-mass, 0.5g of grounded and well mixed plant materials were digested in a diacid mixture of nitric and perchloric acid (4:1). After digestion, the volume was made to 25ml with double distilled water, filtered and stored in well washed plastic bottles and analysed for Ni using the atomic absorption spectrophotometer (Lindsay and Norvell, 1978).

**Desorption Studies:** To study the desorption of Ni from contaminated soil by different chelating agents. Two soil samples for Ni, viz. 1) Ni spiked @ 150 mg Ni $kg^{-1}$ soil. 2) Ni spiked and FYM amended @ 3per cent of soil. Procedure:

**Statistical analysis of data** Factorial CRD for two factors were employed to study the effect of different treatments in various experiments. The analysis was carried out with the help of computer. The effects of treatments were compared with the help of the interaction CD.

## RESULTS AND DISCUSSION

### Tolerance of Ni by Indian mustard genotypes from Ni enriched soil

### Toxicity symptom

To study the tolerance of Ni by Indian mustard three genotypes, RH-819, Varuna and RH-9304 were grown in pots filled with sandy soil spiked with 0, 100, 150, 200 and 250 mg Ni $Kg^{-1}$ soil Ni levels. Visual toxicity symptoms due to Ni were recorded from germination to harvesting of Indian mustard plants. The seeds in all pots were germinated but at $Ni_{250}$ no seedling was grown further. However, the development of plants in control and $Ni_{100}$ were not much differed (Photo plate 1). The leaves were narrow and small with stunted growth, poor branching, delayed flowering and poor seed development and resulted into low biomass yield.

  

Even Ni is considered as an essential micronutrient for plants, but is strongly phytotoxic at higher concentrations (Boominathan and Doran, 2002). However, when present in excess, they can have deleterious effects because of their reactive nature (Schutzendubel and Polle, 2002).

Ni-induced deactivations of proteins including antioxidant enzymes, lipid peroxidation and membrane function have been reported in plants (Madhava Rao and Sresty, 2000).

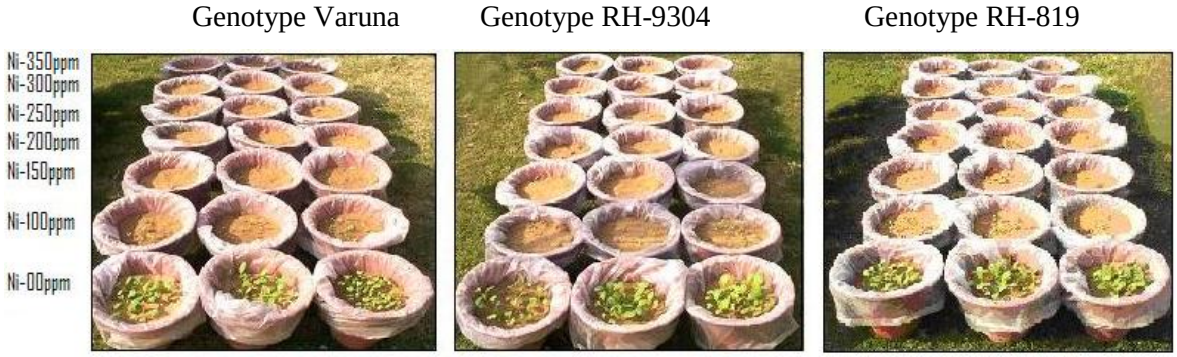


**Photo plate- 1: Effect of different Ni levels on germination of Indian Mustard genotypes**

## Chlorophyll contents

The data in Table 1 showed that Ni application significantly and progressively decreased the mean chlorophyll content 'a' in Indian mustard genotypes from 11.29 mg $g^{-1}$ in $Ni_0$ to 4.78 mg $g^{-1}$ in $Ni_{200}$ treatment. The chlorophyll content 'a' decreased from 13.25 – 6.24, 11.15 – 4.15 and 9.46– 3.95 mg $g^{-1}$ in Indian mustard genotypes of RH-819, Varuna and RH-9304, respectively as the Ni levels increased from 0 – 200 mg Ni $kg^{-1}$ soil.

**Table 1 Effect of Ni application on Chlorophyll content (a, b and total) of Indian mustard genotypes (mg $g^{-1}$) on fresh weight basis**

| Ni levels (mg/Kg soil) | Chlorophyll content (Mg/g) | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Chlorophyll content 'a' | | | | Chlorophyll content 'b' | | | | Total Chlorophyll content | | | |
| | RH 819 | Varuna | RH-9304 | Mean | RH 819 | Varuna | RH-9304 | Mean | RH 819 | Varuna | RH-9304 | Mean |
| **0** | 13.25 | 11.15 | 9.46 | 11.29 | 4.36 | 3.94 | 3.20 | 3.83 | 17.61 | 15.09 | 12.66 | 15.12 |
| **100** | 12.74 | 11.06 | 8.64 | 10.32 | 4.20 | 3.75 | 2.95 | 3.63 | 16.94 | 14.81 | 11.59 | 14.45 |
| **150** | 10.21 | 9.34 | 7.15 | 8.9 | 3.65 | 3.1 | 2.48 | 3.08 | 13.86 | 12.44 | 9.63 | 11.98 |
| **200** | 6.24 | 4.15 | 3.95 | 4.78 | 2.15 | 1.54 | 1.20 | 1.63 | 8.39 | 5.69 | 5.15 | 6.41 |
| **Mean** | 10.61 | 8.92 | 7.30 | 8.94 | 3.59 | 3.08 | 2.46 | 3.04 | 14.20 | 12.01 | 9.76 | 11.99 |
| CD (P=0.05) | Genotype-0.28 | Ni Levels-0.33 | G x Ni-0.57 | | Genotype-0.96 | Ni Levels-1.11 | G x Ni-1.92 | | Genotype-0.24 | Ni Levels-0.28 | G x Ni-0.48 | |

ISSN PRINT 2319 1775 Online 2320 7876
 UGC CARE Listed ( Group -I) Journal Volume 12, Iss 1, 2023

The average chlorophyll content 'a' in Indian mustard plant was 11.29, 10.32, 8.90 and 4.78 mg $g^{-1}$ with 0, 100, 150 and 200 mg $kg^{-1}$ soil Ni applied, respectively. However, the magnitude of decrease in chlorophyll content 'a' varied with the genotypes. The interaction effect between Ni levels and Indian mustard genotypes was significant.

The data on chlorophyll 'b' content of Indian mustard genotypes as affected by Ni levels. It was similar to chlorophyll 'a'content i.e. affected significantly by Indian mustard genotypes and Ni levels in soil. The average chlorophyll 'b' content in plant was 3.83, 3.63, 3.08 and 1.63 mg $g^{-1}$ with the application of 0, 100, 150 and 200 Ni $kg^{-1}$ soil, respectively. The magnitude of chlorophyll content 'b' varied with genotypes. It decreased from 4.36 –2.15, 3.94 –1.54 and 3.20–1.20 mg $g^{-1}$ of RH-819, Varuna and RH-9304 genotypes, respectively with the increase of soil Ni concentration from 0–200 mg $kg^{-1}$ soil.

A perusal of data revealed that the total chlorophyll content in Indian mustard genotypes affected both by Ni content in soil and Indian mustard genotypes. The total chlorophyll content decreased from 17.61 – 8.39, 15.09 – 5.69 and 12.66 – 5.15 mg $g^{-1}$ in Indian mustard genotypes RH 819, Varuna and RH-9304, respectively as the Ni levels increased from 0 – 200 mg $kg^{-1}$ soil. The mean total chlorophyll content of genotypes RH-819 was highest 14.20 mg $g^{-1}$ followed by genotype Varuna (12.01 mg $g^{-1}$) and RH-9304 (9.76 mg $g^{-1}$). The average total chlorophyll content in Indian mustard plant was 15.12, 14.45, 11.98 and 6.41 mg $g^{-1}$ with 0, 100, 150 and 200 mg $kg^{-1}$ soil Ni applied, respectively.

In all studied genotypes the highest values of total chlorophyll content was recorded (17.61 mg $g^{-1}$) at $Ni_0$ in RH-819, whereas the minimum total chlorophyll content (5.15 mg $g^{-1}$) was recorded at $Ni_{200}$ in RH-9304. However, the magnitude of decrease in total chlorophyll content varied with the genotypes. The interaction effect between Ni levels and Indian mustard genotypes was significant with respect to total chlorophyll content in Indian mustard plant.

The reduction in chlorophyll content with application of Ni in soil might be due to decrease in biosynthesis of chlorophyll and iron concentration in Groundnut plant (Nagajyoti *et al.*, 2008) or by induction of its degradation catalyzed by increased chlorophyllase activity. Ni affects electron transport and may cause its complete inactivation (Baier and Dietz, 1999). Moreover, the central metal atom of chlorophyll, Mg, can be substituted with Cd or Ni and ulitmatly prevents light harvesting by the affected chlorophyll molecules (Mishra and Dubey, 2005).

**Dry matter yield**

  

Data on shoot dry matter yield of Indian mustard genotypes from Ni enriched soil are presented in Table 2. It is evident from the data that Indian mustard genotypes showed variability in shoot yield irrespective of different levels of Ni application. The mean dry matter yield of shoot was 28.44, 26.85, 14.61 and 2.54 g $pot^{-1}$ with application of 0, 100, 150 and 200 mg Ni $Kg^{-1}$ soil, respectively. Similar to Cd the shoot dry matter yield of Indian mustard was also influenced by different levels of Ni application. As the Ni concentration in soil increased from 0 to 200 mg $Kg^{-1}$ soil, the mean shoot dry matter yield decreased significantly. The shoot dry matter yield of RH-819 (32.28 g $pot^{-1}$) and RH-9304 (25.05 g $pot^{-1}$) was highest at $Ni_0$ level, while in Varuna genotype it was 28.04 g $pot^{-1}$ at $Ni_{100}$ level. Likewise Parida *et al*. (2003) in a screen house experiment using an alkaline sandy loam soil equilibrated with graded level of Ni (0-300 mg $kg^{-1}$ soil) found an increase in green as well as the dry matter yield of fenugreek upto 21 g Ni $kg^{-1}$ soil but decreased significantly at 40 mg Ni $kg^{-1}$ soil. The dry matter yield of shoot of RH-819, Varuna and RH-9304 ranged from 32.28 to 4.20, 27.98 to 2.25 and 25.05 to 1.17 g $pot^{-1}$, respectively with increase of Ni from 0 to 200 mg $Kg^{-1}$ soil. The maximum mean dry matter yield of shoot was recorded in RH-819 (20.13 g $pot^{-1}$) followed by Varuna (18.31 g $pot^{-1}$) and RH 9304 (15.89 g $pot^{-1}$). The data further showed that the interaction between Ni levels and genotypes was significant. According to Kopittke *et al.* (2007) Nickel was found to be quite toxic to the growth of cowpea, a 10 per cent reduction in relative shoot and root mass occurring at a $Ni^{2+}$ activity of 1.4 µM.

**Table 2 Dry matter yield (g $pot^{-1}$) of different genotypes of Indian mustard as affected by Ni levels**

| Genotypes | Ni levels (mg$Kg^{-1}$ soil) | | | | |
|---|---|---|---|---|---|
| | 0 | 100 | 150 | 200 | Mean |
| **RH 819** | 32.28 | 28.65 | 15.37 | 4.20 | 20.13 |
| **Varuna** | 27.98 | 28.04 | 14.98 | 2.25 | 18.31 |
| **RH-9304** | 25.05 | 23.86 | 13.47 | 1.17 | 15.89 |
| **Mean** | 28.44 | 26.85 | 14.61 | 2.54 | 18.11 |

CD (P=0.05)-; Genotype-0.96; Ni Levels-1.11; G x Ni-1.92;

**Nickel concentration**

The data in Table 3 revealed that the Ni concentration in Indian mustard shoot varied markedly with genotypes and increased significantly with increasing rate of Ni in soil. Applications of graded levels of Ni from 0-200 mg $kg^{-1}$ soil, increased Ni concentration from 0.02 to 67.49, 0.01 to 59.27 and 0.02 to 45.65 µg $g^{-1}$ in RH-819, Varuna and RH-9304 Indian mustard genotypes, respectively and the mean value of Ni content in shoot was 0.016, 31.18, 46.59 and 57.47 µg $g^{-1}$ in control, $Ni_{100}$, $Ni_{150}$ and $Ni_{200}$ treated pots, respectively. These results indicated that shoot Ni concentration significantly and progressively

increased with increasing additions of Ni. The interaction between genotype X Ni was found to be significant.

The mean Ni concentration in shoot of RH-819 was always significantly high (39.19 µg $g^{-1}$) than Varuna (35.15µg $g^{-1}$) and RH-9304 (27.10 µg $g^{-1}$). In shoot of RH 819 it was 11.49 per cent higher than Varuna and 44.61 per cent higher than RH-9304. The maximum Ni shoot concentration was observed in RH-819 at $Ni_{200}$.

**Table 3 Ni concentration (µg $g^{-1}$) in different genotypes of Indian mustard as affected by Ni levels**

| Genotypes | Ni levels (mgKg$^{-1}$ soil) | | | | |
|---|---|---|---|---|---|
| | **0** | **100** | **150** | **200** | **Mean** |
| **RH 819** | 0.02 | 34.62 | 54.64 | 67.49 | 39.19 |
| **Varuna** | 0.01 | 32.65 | 48.65 | 59.27 | 35.15 |
| **RH-9304** | 0.02 | 26.26 | 36.48 | 45.65 | 27.10 |
| **Mean** | 0.016 | 31.18 | 46.59 | 57.47 | 33.81 |

CD (P=0.05):- Genotype-0.24; Ni Levels-0.28; G x Ni-0.48;

**Table 4 Ni uptake (µg pot$^{-1}$) by different genotypes of Indian mustard as affected by Ni levels**

| Genotypes | Ni levels (mgKg$^{-1}$ soil) | | | | |
|---|---|---|---|---|---|
| | **0** | **100** | **150** | **200** | **Mean** |
| **RH 819** | 0.65 | 991.86 | 839.82 | 283.46 | 528.95 |
| **Varuna** | 0.28 | 915.51 | 728.78 | 133.36 | 444.48 |
| **RH-9304** | 0.50 | 626.56 | 491.39 | 53.41 | 292.97 |
| **Mean** | 0.48 | 844.64 | 686.66 | 156.74 | 422.13 |

CD (P=0.05):- Genotype-0.58; Ni Levels-0.67; G x Ni-1.16

**Nickel uptake**

A perusal of data in Table 4 revealed that Ni uptake in shoot increased with increasing levels of Ni application upto 100 mg Ni $kg^{-1}$ soil and thereafter, decreased in all Indian mustard. The results indicated that the mean Ni uptake by shoot was significantly more in RH-819(528.95 µg pot$^{-1}$) as compared to Varuna (444.48 µg pot$^{-1}$) and RH-9304 (292.97 µg pot$^{-1}$). The maximum Ni uptake by shoot was recorded 991.86 µg pot$^{-1}$ in RH-819 at $Ni_{100}$ treatment. The interaction between Genotype X Ni was found to be significant. The Ni uptake by shoot of RH-819 differed significantly from Varuna and RH-9304 at each level of added Ni. Decrease in Ni uptake could be attributed to its less dry matter yield. Similar results were also reported by Gupta *et al.* (2002) in *Brassica juncea*, and Singh and Nayyar

(1998) in forage crops. Purakayastha, *et al.* (2008) found that *B. carinata* showed the highest concentration (mg $kg^{-1}$) as well as uptake (µg $pot^{-1}$) of Ni and Pb at maturity due to its greater biomass production.

## CONCLUSION

The results of this study showed that all the three Indian mustard genotypes survived upto Ni 200 mg $kg^{-1}$ soil level. The genotypes exhibited visual Ni toxicity symptoms with narrow and small leaves, stunted growth, poor branching, delayed flowering and poor seed development and resulted into low biomass yield. Genotype RH-819 produced highest mean biomass, Ni concentration and its uptake on Ni spiked soil. Hence, genotype RH-819 of Indian mustard was found the best Ni tolerant genotype and was selected for uptake study.

## REFERENCE

Ahmed, K.S., Panwar, B.S. and Gupta, S.P. 2001. Phytoremediation of cadmium-contaminated soil by *Brassica* species. *Acta Agronomica Hungarica.* 49, 352-360.

Al-Hawari, A. and Mulligan, C.N. 2006. Heavy metals uptake mechanisms in a fixed column by calcium-treated anaerobic biomass. *Process Biochemistry*. 41(1), 187-198.

Baier, M and Dietz, K.J. 1999. The costs and benefits of oxygen for photosynthesixing plant cells. *Prog. Bot.* 60, 282-314.

Bishnoi, N.R., Dua, A., Gupta, V.K. and Sawhney, S.K. 1993b. Effect of chromium on seed germination, seedling growth and yield of peas. *Agric. Eco. Envion.* 47(1), 47-57.

Cooper, E.M., Sims, J.T., Cunningham, S.D., Huang, J.W. and Berti, W.R. 1999. Chelate assisted phytoextraction of lead from contaminated soils. *J. Environ. Qual*. 28, 1709-1719.

Gupta, S.K., Chaudhary, M.L. and Das, S.N. 2002. Effect of nickel and nitrogen on seed yield, oil content and oil quality of raya (*Brassica juncea* L.) in typic Ustochrept and typic Haplaquept of Haryana. *J. Indian Soc Soil Sci*. 50, 144-146.

Kopittke, P.M., Asher, C.J, Blamey, F.P.C., Menzies, N.W. 2007. Toxic effects of $Pb^{2+}$ on the growth and mineral nutrition of signal grass (Brachiaria decumbens) and Rhodes grass (Chloris gayana). *Pl. Soil*. 300(1-2), 127-136.

Mishra, S. and Dubey, R.S. 2005. Heavy metal toxicity induced alterations in phytosynthetic metabolism in plants. In *Hand Book of Phytosynthesis 2nd Ed*. By Mohammad Pessarakli. CRC Press. 845-864.

Nagajyoti, P.C., Dinakar, N., Prasad, T.N.V.K.V., Suresh, C. and Damodharam, T. 2008. Heavy metal toxicity: Industrial Effluent Effect on Groundnut (*Arachis hypogaea* L.) Seedlings. *J. App. Sci. Res.* 4(1), 110-121.

Okoronkwo, N.E., Ano, A.O. and Onwuchekwa, E.C. 2005. Environment, health and risk assessment: a case study of the use of an abandoned municipal waste. *Am. J. Biotechnol.* 4(11), 1217-1221.

Purakayastha, T. J., Viswanath, T., Bhadraray, S., Chhonkar, P.K., P. P. Adhikari, P.P. and Suribabu, K. 2008. Phytoextraction of Zinc, Copper, Nickel and Lead from a Contaminated Soil by Different Species of *Brassica*. *Int. J. Phyto.* 10(1), 61–72.