



## Effect of Nickel ion on Stem of *Hydrilla Verticillata* L.

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### Abstract

The study sought to evaluate the potential of *Hydrillaverticillata* L. in the absorption of Nickel(Ni) ion and possible variations in its tissues after 7 days of exposure to this metal. *Hydrillaverticillata* L. were cultured in Hoagland medium supplemented with various Ni ion concentrations (as 3,5,7,9 and 11 mg/ml) and were separately harvested after 3, 5 & 7 days. In the anatomical analysis, disorganization of epidermal cells, degeneration of cortical cells and pith, highlights the variation resulting from Ni ion toxicity. However these variations were not sufficient to damage the development of an individual. *Hydrillaverticillata* L. showed high capacity of extraction and storage of the metal, being food alternative to aquatic environments, with high concentration of Ni ion.

**Keywords:** Anatomical variations, aquatic environment, hoagland medium, *Hydrillaverticillata* L., Ni ion toxicity, stem

### Introduction

Heavy metals at high concentration in substrate when taken up by the plants develop toxic characters, which becomes expressed with anatomical alterations or even malformations. Although some heavy metals are essential trace elements for plant life, at relatively high concentrations they are toxic since they interfere with enzyme function (Krupa *et al.*, 1993). Nickel has many visible and adverse effects on environment. The foremost adverse effect is Skin allergy. The other detrimental effects are Nickel compounds are carcinogenic as well as cause asthma. Hence, its removal is of major concern. Moreover, very little research has been conducted on the mechanisms of Ni phytotoxicity. In general, heavy metals severely inhibit root growth (Bennet, 1987 and Punz, 1993). Furthermore, several studies have indicated that Ni inhibits plant photosynthesis (Bishnoi, *et al.* 1993, Clijsters and Assche 1985. and Krupa *et al.* 1993). Bio availability of heavy metal in soil, uptake of heavy metal at phytotoxic level, growth retardation,

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effects on palisade and spongy parenchyma cells in leaves (Ahmed, 2003 and Ladygein and Sharma 2004) collated deposition in the vascular bundles and change in vacuoles with electron dense material along the walls of xylem and phloem vessel (Ladygein and Semenova, 2003). Therefore, in the present study, the effects of high Ni ion concentrations on the stem of *Hydrillaverticillata* L. were studied in order to determine the structural features of Ni ion toxicity and their potential physiological implications in response to Ni toxicity.

### Material and Methods

#### Plant Material

*Hydrillaverticillata* L. plants were collected from the pond at Harani, Vadodara (Fig. A). They were allowed to acclimatize for 15 days. Plants were grown and propagated for 4 weeks in quarter strength Hoagland's solution (Hoagland and Arnon 1938). In the pilot scale experiment, after determining LC 50 value mg/ml 254 hours, the test plants were exposed to wide range of the metal ion concentrations i.e. 3, 5, 7, 9 and 11 mg/ml. Nutrient solution devoid of trace element served as a control. Both the control and the treated solutions were maintained at pH 5.5 using dilute HCl or NaOH. After each experimental period, harvested plants



were washed in running tap water and rinsed with deionized water.

### Microscopy

To observe anatomical changes in Ni exposed cells of *Hydrillaverticillata*L. following technique was used: The control and 11 ppm cd treated cells of the test plants were preceded for microtechnique (Johansen, 1940) method. Measurements and photographs were taken using a Leica DM1000 binocular light microscope with a Leica DFC280 camera. Observations were made on organization of epidermal cells, cells of cortical layer and central cylinder (pith) in the control and treated cells of *Hydrillaverticillata*L.

### Results and Discussion

Anatomical studies on *Hydrillaverticillata* L plants showed that the stem in cross section exhibits an oval profile (Fig C). On examination of control stem of *Hydrillaverticillata* L revealed that uniformly distributed radially narrow epidermal cells (Fig. B), no conspicuous cuticle over epidermis, well organized cells of cortical layer with compactly arranged parenchyma cell interrupted by arenchyma cells (Fig. C). At the center of the central cylinder was a large lecuna (Fig. D). The stem of *Hydrillaverticillata* L.

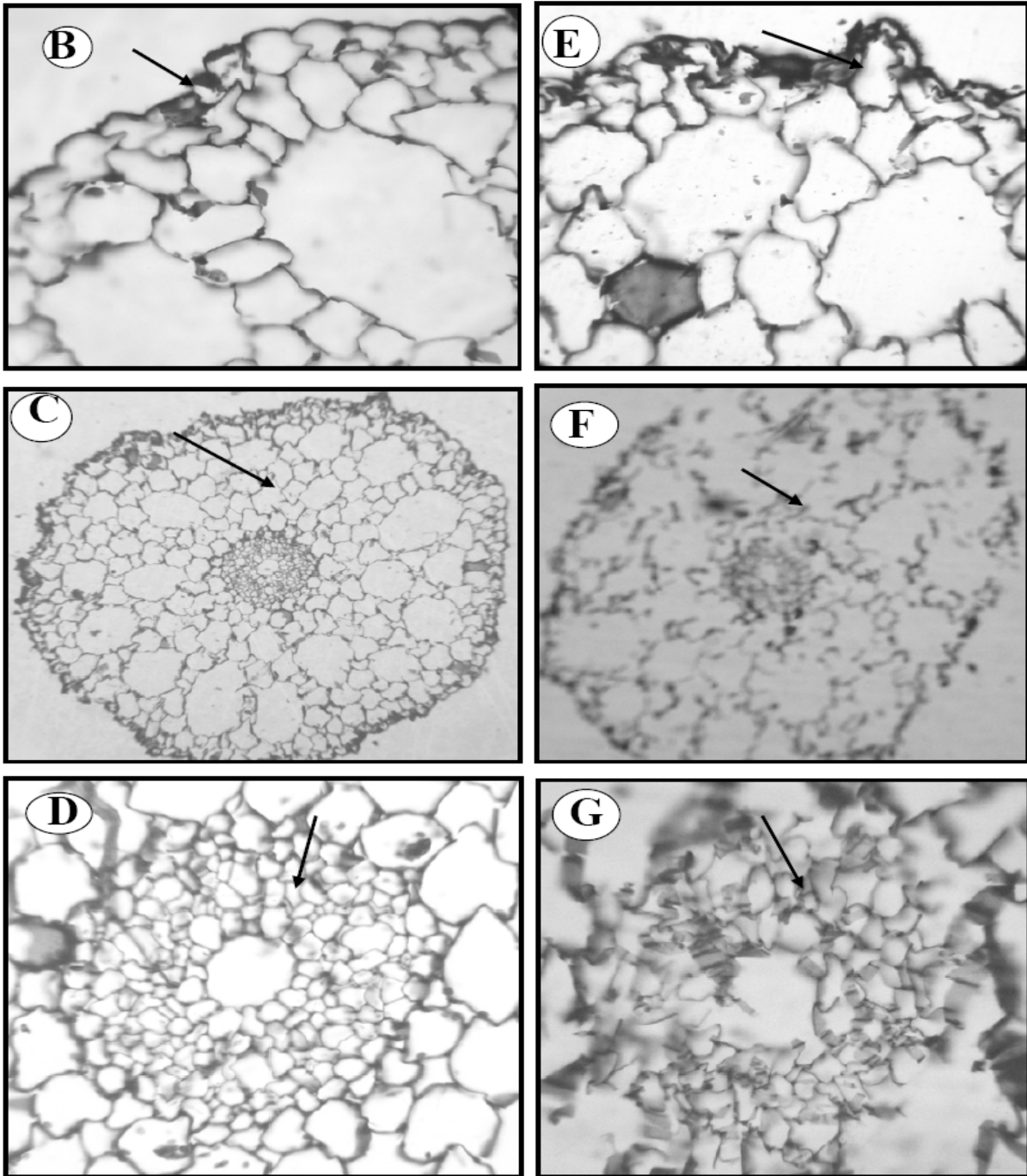
growing in excess of Nickle ion exhibits anatomically number of differences compared to the control stem (Fig.F).

Thus, the stem profile is much larger, a fact principally due to the increase of the volume of pith. Disturbance in arrangement of epidermal cells (Fig. E), cortical cells are disintegrated forming a dark zone (Fig. F) and pith does not remain any longer compact, but its concentration region becomes disorganized resulting in an open cavity (Fig. G) were major observation in Ni treated cells. In the present study, anatomical studies on stem cross sections of *Hydrillaverticillata* L. revealed that the histological components which appeared significantly affected by Ni toxicity were the epidermis, cortex, and pith. The relative volume of the cortex became reduced in treated plant as compare to control due to disorganization of the parenchyma tissues (Panou-filotheouet *al.* 2006 Sridhar, *et al.*, 2007). The volume of pith region increased through the development of larger central cavity formed by tearing apart of the pith cells. (Panou-filotheouet *al.* 2006).

The formation of dense pubescence in the stems of oregano plants grown in high Cu-concentrations is rather attributed to the stress conditions developed by Cu toxicity, as in other relevant cases (Barceloet *al.*, 1988 and Panou-Filotheouet *al.* 2001).



**Fig. A: Collection site**



**Fig. B: Intact outer epidermis**

**Fig. C: Transverse section of stem of control plant**

**Fig. D: Pith of control plant**

**Fig. E: Disorganization in cells of epidermis**

**Fig. F: Degeneration of parenchyma cells of cortical layer**

**Fig. G: Disintegration of pith cells.**

### Effect of Nickel ion on Stem

Considering the specific alterations (under Ni stress) in *Hydrillaverticillata* L. stem structure and their evaluation by morphometric assessments, it could be suggested that increasing Ni concentrations have a toxic effect on stem. This effect becomes anatomically expressed by a disorganization of a great amount of parenchymatic tissue in the stem cortex and pith. Additional physiological studies (endogenous gibberellic acid and other phytohormones, phenoloxidase and other enzymes, saps of vessels and sieve tubes, etc.) would strengthen structural data and provide grounded interpretations as to the manner of toxic action of Ni.

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