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Short Communication

Phytoremediative Potential of Tobacco under Deficit Irrigation Conditions for Ni-Polluted Soil

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Abstract

The aim of this study was to investigate the possibility of reclaiming Ni-polluted soils by phytoremediation under deficit irrigation conditions. Tobacco, *Nicotiana rustica*, was grown for 6 weeks at four different irrigation rates (100, 80, 60, and 40% of irrigation requirement) and four nickel concentrations (0, 50, 100, and 200 mg Ni·kg^{-1} soil) applied from $NiSO_4$. Nickel toxicity, water stress symptoms, dry shoot biomass, Ni concentration, and Ni uptake were followed. Neither Ni treatment nor water stress-induced Ni toxicity were observed, and there was no treatment-induced difference in chlorophyll content of leaves. With the increasing Ni application, there was a gradual increase in Ni concentration of the shoots from the 40% irrigation through 100% irrigation. As a result, the ability of tobacco to accumulate Ni at high concentration can enable it to be used for phytoremediation of Ni-polluted soils despite the fact that water deficit limits Ni accumulation to some extent.

Keywords: phytoremediation, irrigation, nickel, tobacco, water stress

Introduction

Soil and water are two essential natural resources for life cycles. Both limited freshwater resources and increasing quality-related problems have resulted in water scarcity as a serious problem in the world. Thus, different strategies should be developed to overcome the deleterious water scarcity problem. Deficit irrigation could be one strategy, which may be defined as to set an economically and environmentally sound balance between available freshwater resources and drought hazard.

Soil degradation and pollution seriously limits the ability of soil to support natural lifecycles. Nickel (Ni), one of the heavy metals, is a common soil pollutant. Despite Ni being an essential nutrient required in very small amounts, its high concentrations are toxic to plants, animals, and humans [1-3]. Nickel concentration in soil varies from 3 to 1,000 mg·kg^{-1} [2], but it may reach as high as 5,000 mg·kg^{-1} in soils formed on ultra basic volcanic parent materials rich

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Table 1. The effect of Ni concentration on dry biomass, chlorophyll content of young and old leaves, Ni concentration of shoots, and amount of Ni uptake by shoots.

Ni treatments ($mg \cdot kg^{-1}$)	Dry weight ($g \cdot plant^{-1}$)	Chrolophyll (SPAD reading)		Ni concentration ($mg \cdot kg^{-1}$)	Ni content ($\mu g \cdot plant^{-1}$)
		Old leaves	Young leaves		
0	12.2 b	40.0 a	52.8 b	9.46 d	115 c
50	13.0 a	37.7 ab	55.6 a	36.5 c	465 b
100	12.8 a	36.1 b	55.8 a	44.4 b	572 a
200	11.2 c	38.5 ab	54.6 ab	57.2 a	613 a

Different letters indicate significant differences between the means.

in serpentine minerals [4]. On the other hand, anthropogenic activities, mining, agriculture, and industrial practices, fossil fuel usage, etc., may be listed as the main cause for increasing the pollution load of agricultural soils [2, 5].

Nickel-polluted soils may be remediated through various chemical, physical, and biological methods. The phytoremediation method, a biological method, has drawn attention in recent years because it is easy to apply, economically affordable, and environmentally- and aesthetically-friendly. Phytoremediation may be defined as the uptake of pollutant agents such as heavy metals by roots and then transportation and accumulation of the pollutants in above-ground parts of the plants [6]. The plants to be used for phytoremediation should produce abundant shoots, deep roots, be fast growing, easily harvestable, and accumulate high amounts of heavy metals with high tolerance to high concentrations of the metals [7]. There have been many reports in literature in the last decade dealing with the phytoremediation of different heavy metals [7-11].

In this study, the remediation possibility of Ni-polluted soils under deficit irrigation growth conditions was investigated. Tobacco is selected as the phytoremediator because of its common usage for phytoremediation, non-selective nature for climate and soil, deep roots, and abundant shoot-producting ability [7-8].

Experimental Procedures

Nicotiana rustica (Tömbeki tobacco) was grown in pots filled with surface samples (0-30 cm) of Paşaköy soil series. The properties of soil were as follows: loam texture [12], pH 7.83 [13], $CaCO_3$ 5.22% [14], organic matter 1.58% [15], and DTPA-extractable Ni 7.49 $mg \cdot kg^{-1}$ [16].

Three kg of soil was filled in 4 L plastic pots. The following amounts of fertilizers were applied to each pot: 200 $mg \cdot kg^{-1}$ N as $(NH_4)_2SO_4$ form, 100 mg $P \cdot kg^{-1}$ and 125 mg $K \cdot kg^{-1}$ as KH_2PO_4 form, and 2.5 $mg \cdot kg^{-1}$ Fe as FeEDTA. Ni concentrations of 0, 50, 100, and 200 $mg \cdot kg^{-1}$ as $NiSO_4$ were applied to the soil in solution and immediately irrigated to 90% of field capacity for homogenous distribution of the added metal. The pots were then incubated for 3 weeks at 25ºC to allow short-term adsorption reactions. Pre-germinated seedlings were transformed to the plots. When the surface coverage of the plant reached 20-30%, irrigation deficit regimes (irrigation rate (IR) IR_{40}, IR_{60}, IR_{80}, and IR_{100}) were started. At first, the pots irrigated to field capacity and then before the subsequent irrigation pots were weighed and the difference between field capacity and their current weights were used to calculate the irrigation requirement for IR_{100} treatment. The amount of irrigation water for IR_{80}, IR_{60}, and IR_{40} treatments were 80, 60, and 40 percent of IR_{100} treatment, respectively. Irrigation was repeated in approximately 3-day intervals depending on the appearance of the plants in IR_{100} treatment.

Tobacco seeds were germinated in a mixture of peat and sand (2:1, V/V). *Nicotiana rustica* tobacco plants were grown under controlled environmental conditions with a 16 h light period (light intensity of 10 klux), a 25/20ºC light/dark temperature regime, and 60% relative humidity. Chlorophyll content of leaves was determined using a chlorophyll meter (Konica-Minolta Spad-502) before harvesting the plants. Plants were harvested after 6 weeks of growth. The samples were rinsed briefly in deionized water and dried with tissue paper, then dried at 70ºC. After shoot dry weights were measured, the particle size was reduced with an agate mill (Retsch RM200). Then the samples were wet combusted with HNO_3 and H_2O_2 in a microwave oven (MarsXpress CEM) and total Ni concentrations of digested samples were measured using an ICP-AES (inductively coupled plasma-atomic emmission spectrometry; Varian Series-II).

The experiment was set up in a completely randomized design with three replications. Analysis of variance was performed using a general linear model procedure and the effect of treatments were compared by LSD test at $p \leq 0.05$ [17].

Results and Discussion

Visual observations showed that there were no toxicity symptoms even at 200 $mg \cdot kg^{-1}$ Ni treatment and at minimum (IR_{40}) irrigation rate. There were significant Ni treatment-induced differences in chlorophyll content of both old and young leaves (Table 1), but irrigation deficit affected only the chlorophyll content of old leaves (Table 2). The chlorophyll content showed an increasing trend from old leaves through young leaves. The occurrence of no treat-

Table 2. The effect of irrigation ratio (water deficit) on dry biomass, chlorophyll content of young and old leaves, Ni concentration of shoots, and amount of Ni uptake by shoots.

Irrigation rate (%)	Dry weight ($g \cdot plant^{-1}$)	Chlorophyll (SPAD reading)		Ni concentration ($mg \cdot kg^{-1}$)	Ni content ($\mu g \cdot plant^{-1}$)
		Old leaves	Young leaves		
100	16.6 a	36.5 b	55.1 a	34.8 b	573 a
80	13.3 b	40.2 a	54.6 a	36.8 ab	464 b
60	11.1 c	37.2 ab	54.6 a	39.7 a	426 bc
40	8.10 d	38.3 ab	54.5 a	36.9 ab	304 d

Different letters indicate significant differences between the means.

ment-induced visual symptoms in tobacco species used in this study suggests that either tobacco is very tolerant of the stress factors or the level of the stress factors is not strong enough to cause visual symptoms. It was reported by Panwar et al. [9] that no visible Ni toxicity symptoms were observed in *Brassica juncea* and *Brassica carinata* plants up to the 80 mg $Ni \cdot kg^{-1}$ treatments in soil but slight decreases in plant height.

Nickel treatments significantly affected dry biomass production (Table 1). There was no significant difference in fresh biomass production (data not given), but a slight decrease was observed for the 200 $mg \cdot kg^{-1}$ treatment. However, with increasing irrigation deficit, both fresh and dry shoot biomass decreased (Table 2). Ni treatments, irrigation deficit treatments, and Ni treatment × IR interactions effects on dry biomass were found to be statistically significant. 50 $mg \cdot kg^{-1}$ Ni treatment significantly increased the dry biomass compared to the control (0 $mg \cdot kg^{-1}$ Ni treatment) from 12.2 to 13.0 $g \cdot pot^{-1}$. The subsequent increasing Ni treatments gradually decreased biomass. Similar to the findings obtained in this study, increasing Ni applications resulted in significant reduction in both dry and fresh shoot biomass [5]. On the other hand, Chen et al. [18] reported a linear decrease corresponding to the increase in Ni concentration.

As Ni concentration of soil increased, the Ni uptake by plants increased from 9.46 to 57.2 $mg \cdot kg^{-1}$ (Table 1). Ni treatments strongly affected the Ni concentration of tobacco shoots (Table 1), whereas the deficit irrigation's effect was so clear despite statistical significance (Table 2). Interaction of the stress factors significantly influenced plant Ni concentration (Data not given). Singh and Pandey [19] reported a decrease in relative water content of *Pistia stratiotes* plants, depending on the increasing Ni treatments (0-10 $mg \cdot kg^{-1}$). Panwar et al. [9] found that *Brassica juncea* is more tolerant and accumulate more Ni than *Brassica carinata* under increasing Ni stress (0, 40, and 80 $mg \cdot kg^{-1}$), and Ni treatment increased plant Ni concentration. Water deficit treatments significantly affected all of the measured parameters ($p \leq 0.001$) except chlorophyll content of young leaves.

As a result, the ability of tobacco to accumulate Ni at high concentration can enable it to be used for phytoremediation of Ni-polluted soils despite the fact that water deficit limits Ni accumulation to some extent.

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