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Effect of Nano Zeolite on the Transformation of Cadmium Speciation and Its Uptake by Tobacco in Cadmium-contaminated Soil

Nano Zeolite impact soil Cd Speciation and Cd Uptake by Tobacco

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Abstract: The study was aimed at exploring the effects of applying ordinary and nano zeolite on the soil pH, soil available cadmium (Cd) content, soil Cd speciation and Cd uptakes by tobacco using pot experiment with simulated Cd contaminated soil indoors. The results showed that soil pH increased and available Cd content reduced with the amount of ordinary and nano zeolite increasing. Compared with the control, the application of ordinary and nano zeolite increased soil pH at 0.47 - 1.05 and 0.73 - 1.57, respectively, and reduced the available Cd contents at 19.3% - 32.7% and 23.2% - 40.5%, respectively. In addition, soil pH had significantly negative correlation with available Cd content in each treatment ($p<0.05$). Nano zeolite could more effectively reduce Cd contents of all parts of tobacco than ordinary zeolite with the same amount treatments, and Cd content in all parts of tobacco plants was positively correlated with soil available Cd content. The content of exchangeable Cd (EXE) in soil decreased to some extent with different zeolite treatments, application of nano zeolite was better than that of application of ordinary zeolite in reducing Cd bioavailability and transferability. Overall, application of nano zeolite has an advantage over ordinary zeolite in reducing available Cd content in soil and Cd content in tobacco.

Keywords: Nano Zeolite; Ordinary Zeolite; Soil Cd speciation; Cd uptake; Tobacco.

1 Introduction

The heavy metal pollution has gained increasing atttention by the public and scientific researchers in recent years [1] . Cadmium (Cd) is one of the most toxic heavy metal elements in soil environment due to its strong chemical activity, large mobility, persistent toxicity, and nonbiodegradability [2,3]. Tobacco is a Cd enriching plant, the accumulation amounts of Cd in tobacco leaves accounted for 50% - 80% of the total Cd absorption amounts [4-5]. Tobacco is a product consumed by inhalation, during which Cd can enter human body through tobacco smoke. There is evidence that majority (approximately 70%) of Cd contained in cigarettes enter tobacco smoke, posing a health hazard to both active and passive smokers [6]. Some reports pointed out that the half-life of cadmium in human body is 13.6 - 23.5 years [7], Cd accumulation amounts of lifelong smokers is 2 times greater than that of non-smokers, and Cd concentration of blood of lifelong smokers is 4-5 times higher than that of non-smokers [8]. High Cd concentrations of blood for a long time is able to cause some organs disease and even cancer [9-10]. Therefore, it is important to explore how to reduce Cd absorption by tobacco through improving soils for tobacco planting.

In situ immobilization of heavy metals in contaminated soils by adding extraneous active amendments has been

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considered as a convenient and low cost measure for contaminated soil remediation. Some immobilization amendments were applied into the heavy metals contaminated soil, such as limestone, zeolite, phosphate and organic material, as well as nano-bio-repair materials and their combinations [11,12], can decrease the available fractions of heavy metals or change their redox states, and thus, effectively decrease the mobility, bioavailability, and toxicity of the heavy metals in soils. Therefore, the method of in-situ immobilization on soil heavy metals contamination has been widely used.

Zeolite is a porous aluminosilicate mineral. It has unique three-dimensional space frame structures, a huge surface area, a great ion adsorption and exchange capacity, which can improve physical and chemical characteristics of soil effectively and increase the utilization ratio of soil nutrient. In addition, the zeolite resources are rich in China, and its total amount is more than 10 billion tons, occupies third of the world resources [13]. Therefore, zeolite has been recognized as the high quality amendments for heavy metals contaminated soil [14,15]. However, the structure and nature of ordinary zeolite has some defects, such as the channels are easily confined, which affects the ion exchange capacity and the surface adsorption capacity, leading to the zeolite showing some limitations in the process of soil Cd pollution repair [14,16]. Nano zeolite after modification possesses the loose skeleton structure, uniform hole structure, larger specific surface area and unique adsorption function [17]. Theoretically, nano zeolite has a larger adsorption abilities and adsorption capacity than ordinary zeolite, which will be a great advantage in the heavy metal cadmium pollution repair for soil. Application of nano zeolite in control of soil Cd pollution and tobacco Cd absorption has not been reported at present. In this study, tobacco pot experiment was carried out to simulate the effects of ordinary zeolite and nano zeolite on soil pH, available Cd content, soil Cd speciation changes and Cd uptakes in tobacco plants. The results of this study provide a cost-effective method for control of heavy metal pollution, and contribute to theoretical basis and technical support for reduction of Cd uptake by tobacco in Cd-contaminated soils.

2 Materials and Methods

2.1 Materials

The tested tobacco cultivar was Yunyan 87 from Hunan Provincial Tobacco Company, Changsha City. The tested soil was the rotation soil (0-20 cm) for tobacco and rice from the Hunan Agricultural University Base of Middle South Agricultural Experimental Station of China Tobacco. The collected soils were air-dried, sieved using 2 mm sieve and placed into plastic bags for future use. The soil physical and chemical properties were measured by a previously reported method [18], and the results were as follows: pH 6.01, total Cd 0.35 mg kg^{-1}, Organic matter 28.5 g kg^{-1}, Alkali-hydrolyzable N 87.64 mg kg^{-1}, Available P 18.45 mg kg^{-1}, Available K 92.03 mg kg^{-1}. The test ordinary zeolite and nano zeolite were purchased from a mine in Henan Province of China. The particle sizes were 0.15 mm and 80-90 nanometers, pH values were 8.8 and 8.5, cation exchange capacities (CEC) were 165 cmol kg^{-1} and 238 cmol kg^{-1}, total specific surface areas were 268.2 m^2 g^{-1} and 572.8 m^2 g^{-1}, the total Cd content were 0.33 mg kg^{-1} and 0.56 mg $kg^{-1,}$ respectively.

2.2 Pot test

The experiment was carried out in the greenhouse of the College of Plant Protection, Hunan Agricultural University from March 26 to June 26, 2017 using plastic buckets with 10.0 kg of the tested soil per pot. The analytical pure $CdCl_2.2.5H_2O$ was used as Cd source and added as solution to the tested soil. The $CdCl_2.2.5H_2O$ solution was mixed well with the soil to reach 10 mg kg^{-1}[19], which is 10 times the Cd standard limits (1.0 mg kg^{-1}) of the Grade III Environmental Quality Standard for Soils of China.

The soils were then treated with tobacco active organic and inorganic base fertilizer (total nutrient ≥ 29%, the ratio of nitrogen, phosphorus and potassium= 8:10:11) with 20, 40 and 60 g kg^{-1}ordinary zeolite (OZ1,OZ2 and OZ3) and nano zeolite (NZ1,NZ2 and NZ3) at room temperature for two weeks prior to transplanting tobacco seedlings. Soil without any treatment was considered as the control (CK). Therefore, there were a total of 7 treatments. Three pots were used for each treatment. Two seedlings were transplanted (at 2 cm depth) into each pot. During the growing stage, special tobacco fertilizer (total nutrient content ≥ 42%, the ratio of nitrogen, phosphorus and potassium is 10:0:32) and agricultural potassium nitrate (total nutrient = 58%, K_2O ≥ 44.5%, and total nitrogen (N) ≥ 13.5%) were applied once. At the maturation stage, the upper, middle and lower leaves and stems of tobacco samples were washed with deionized water and oven-dried at 105°C for 30 min and then 75°C for 48 h. After being dried to constant weight, samples were ground with a stainless steel mill. The rhizosphere soil samples of each plant were collected from roots by shaking soil attached to roots. The rhizosphere soil samples were then transported to the laboratory, naturally air-dried and sequentially sieved using a 100-mesh sieve, and collected for future measurement.

2.3 Measurements and Methods

2.3.1 Determination of soil pH and available Cd content

Soil pH was determined using a pH meter after mixing with water at 1.0:2.5 ratio [18]. Soil available Cd was extracted using diethylenetriaminepentaacetic acid (DTPA) [20]. Soil available Cd concentration in extracting solution was determined by ICP-MS (Thermo Fisher Scientific, USA)

2.3.2 Determination of Cd forms in soil

Chemical speciation of Cd was determined using Tessier's sequential extraction method [21]. Different chemical speciation of Cd was determined by ICP-MS (Thermo Fisher Scientific, USA)

2.3.3 Determination of Cd in tobacco

Tobacco samples (0.2 g, weighed accurately to 0.0001 g) were microwave-digested (CEM, USA) with 8 mL of HNO_3 at 180°C for 30 min. Cd concentration in digestion solution were determined by ICP-MS (Thermo Fisher Scientific, USA)

2.3.4 Data processing and analysis

All data were measured three times and analyzed using Microsoft Excel 2003 software. Statistical analyses and correlation analyses were performed using SPSS 17.0 statistical software.

Ethical approval: The conducted research is not related to either human or animals use.

3 Results and Discussion

3.1 Effects of different zeolite treatments on soil pH

The results showed that application of different zeolite had certain effects on soil pH (Figure 1). Application of

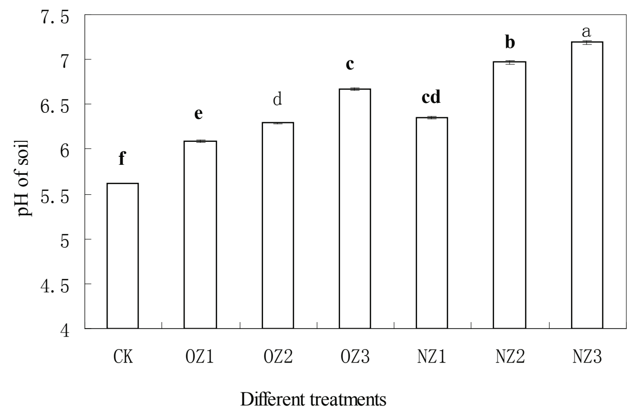

Figure 1: pH of soils in different zeolite treatments (Error bars indicate standard errors of 3 replicates. The different letters indicate significant difference at $P<0.05$, similarly hereinafter).

different amounts of ordinary zeolite and nano zeolite could improve soil pH. Soil pH increases with the increase of the amount of zeolite. Compared with that of control, the pH of soils OZ1, OZ2 and OZ3 treated with ordinary zeolite increased by 0.47, 0.67 and 1.05, respectively. The pH of soils NZ1, NZ2 and NZ3 treated with nano zeolite increased by 0.73, 1.35 and 1.57, respectively. Zeolite can increase soil pH could be explained from two aspects. On the one hand, zeolite itself has a higher pH. Thus, the greater the amount of zeolite used, the greater the increase in pH. On the other hand, zeolite has very strong ion exchange ability. There are a large number of exchangeable K^+, Na^+, Ca^{2+}, Mg^{2+} and other cations. Application of zeolite promotes the exchange of K^+, Na^+ and other soluble cations with H^+ and Al^{3+} in soil, resulting in decline of hydrolyzed and metabolic acids [22]. However, soil pH with nano zeolite treatments had a greater increase than the same amount of ordinary zeolite treatments. This result could account for the fact that nano zeolite has looser skeleton structure, larger specific surface area, larger cation exchange capacities than ordinary zeolite in soil.

3.2 Effects of different zeolite treatments on the available content of Cd in soil

As can be seen from Figure 2, the use of different zeolite can reduce the content of available Cd in the soil, and the more the zeolite was applied, the greater the reduction of available Cd content. Compared with that of the control, available Cd decreased by 19.3% - 32.7% and 23.2% - 40.5% in the soil treated with ordinary zeolite (OZ1-OZ3) and nano zeolite (NZ1-NZ3) , respectively. The former was

Table 1: Effects of ordinary zeolite and nano zeolite on Cd concentrations in various tissues of tobacco plants (Error bars indicate standard errors of 3 replicates. The different letters indicate significant difference at P< 0.05, similarly hereinafter).

Treatments	Upper leaves (mg kg^{-1})	Middle leaves (mg kg^{-1})	Lower leaves (mg kg^{-1})	Stems (mg kg^{-1})
CK	15.35±1.25 a	16.95±2.56 a	19.52±1.89 a	8.01±0.98 a
OZ1	12.32±1.08 b	13.82±1.81 b	15.87±1.66 b	6.23±0.81 b
OZ2	11.58±1.05 bc	12.96±1.38 bc	14.58±1.35 bc	5.85±0.45 bc
OZ3	11.23±0.98 bc	12.35±1.30 bc	14.20±1.20 bc	5.59±0.43 bc
NZ1	11.56±0.95 bc	12.68±1.78 bc	14.49±1.81 bc	5.68±0.70 bc
NZ2	10.89±1.01 c	11.97±1.56 bc	13.85±1.51 c	5.41±0.65 c
NZ3	10.32±1.05 c	11.24±1.30 c	13.20±1.42 c	5.11±0.58 c

significantly lower than the latter with the same amount treatments probably due to that the nano zeolite could more effectively improve soil pH than ordinary zeolite. The results show that nano zeolite could better remediate Cd-contaminated soil than ordinary zeolite.

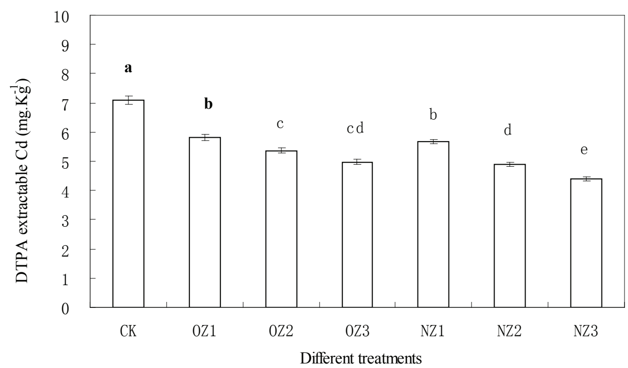

Figure 2: Effects of application of ordinary zeolite and nano zeolite on the content of available Cd in soil.

3.3 Effects of different zeolite treatments on Cd absorption in different parts of tobacco

Table 1 shows the distribution of Cd in different parts of tobacco. The order of Cd content in tobacco is lower leaves> middle leaves> upper leaves> stems. Application of ordinary and nano zeolite could effectively reduce Cd contents in all parts of tobacco. Compared with control, application of ordinary zeolite reduced Cd content by 19.7% - 26.8%, 18.6% - 27.9%, 18.7% - 27.9% and 22.2% - 30.2% in upper, middle and lower leaves and stems of tobacco, respectively; application of nano zeolite reduced Cd content by 24.7% - 32.8%, 25.3% - 33.8%, 25.8% - 32.4% and 29.1% - 36.2% in upper, middle and lower leaves and stems of tobacco, respectively. The results show that nano zeolite could more effectively reduce Cd contents of all parts of tobacco than ordinary zeolite with the same amount of treatments.

3.4 Correlation of soil available Cd content with soil pH and Cd content in different parts of tobacco

The correlation between soil pH under different treatments and cadmium content in different parts of tobacco with available Cd content in soils were analyzed. The results are shown in Figures 3 and 4. It was found that soil pH was negatively correlated with the content of available Cd ($n=7$, $p<0.05$). With the increase of soil pH, the content of available Cd decreased, indicating that pH is one of the important factors affecting available Cd content. Consistent with past research [23], the content of available Cd in different soils was significantly correlated with Cd content in upper, middle and lower leaves and stems of tobacco ($n=7$, $p<0.05$). The application of nano zeolite and ordinary zeolite can reduce soil available Cd content, so as to reduce the absorption of soil Cd in various parts of tobacco.

3.5 Effects of different zeolite treatments on Cd speciation in soil

The Tessier's sequential extraction method [21] is one of the most significant methods for analyzing the forms of heavy metals in soil and sediments and has been widely used in the analysis of soil heavy metals and their toxicity and bioavailability [24-26]. In this method, heavy metals are divided into five forms, namely, exchangeable (EXE),

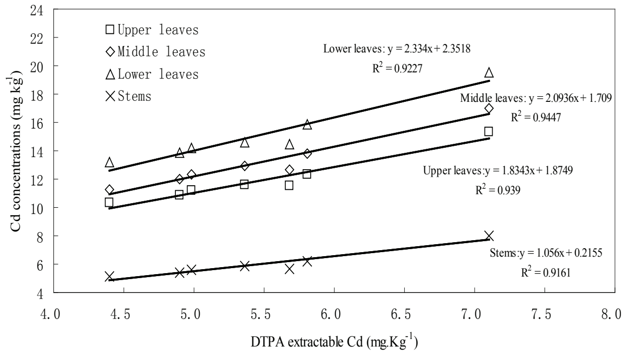

Figure 3: Relationship between pH of soils with different zeolite treatments and soil available cadmium content.

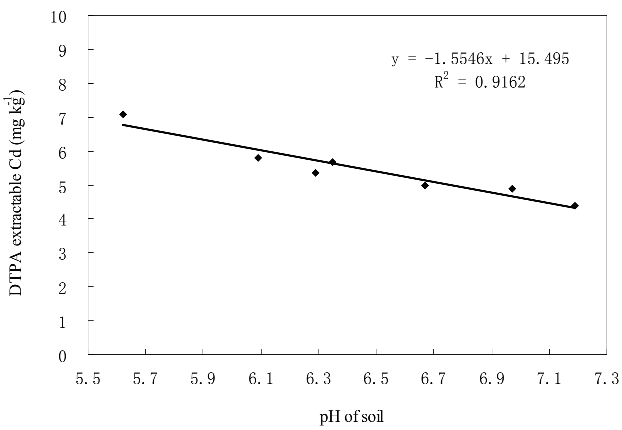

Figure 4: Relationship between soil available cadmium content and cadmium concentrations in different tissues of tobacco plants.

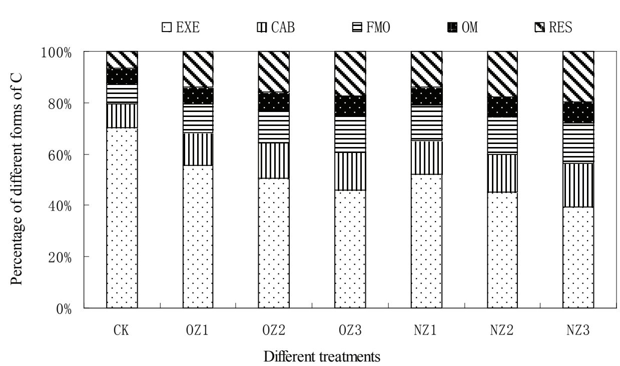

Figure 5: Effects of application of ordinary zeolite and nano zeolite on percentage of different speciation of cadmium in soil.

carbonates-bound (CAB), organic matter bound (OM), Fe-Mn oxides bound (FMO) and residual (RES) forms.

Figure 5 shows the effect of different zeolite treatments on the transformation of Cd speciation in the soil. It can be seen from the figure that the Cd in the untreated soil CK is mainly in the EXE form, accounting for 70.5%, followed in turn by CAB (9.1%), FMO (8.3%), RES (6.5%) and OM (5.7%) of the total Cd ratio. In the soil, soil exogenous Cd was changed toward CAB, FMO, OM and RES under the action of ferromanganese di(tri)basic oxide colloid, salty ion and organic matters, indicating that soil itself has a certain self-purification capacity [27]. Water-soluble and EXE heavy metals are defined as available according to their bioavailability and can be easily absorbed by plants. Metals in FMO, CAB and OM forms are defined as potentially available and the direct provider of available heavy metals. Heavy metals in RES form are defined as unavailable. In the soil-plant system, the order of Cd bioavailability in each form is EXE>CAB>OM>FMO>RES [28]. The speciation of heavy metal Cd in soil changed significantly after applying different zeolite to the polluted soils. Application of different dosages of ordinary zeolite reduced EXE Cd by 14.9% - 24.5% while CAB Cd percentage increased by 3.5% - 5.5%, OM cadmium percentage increased by 0.8% - 1.9%, FMO Cd percentage increased by 3.2% - 6.3% and RES Cd percentage increased by 7.3% - 10.7%, respectively. Application of different dosages of nano zeolite reduced EXE Cd by 18.4% - 31.3% while CAB Cd percentage increased by 4.1% - 8.2%, OM cadmium percentage increased by 1.2% - 2.4%, FMO Cd percentage increased by 5.6% - 7.3% and RES Cd percentage increased by 7.4% - 13.3%, respectively. It can be seen that the effect of nano zeolite treatments is better than that of ordinary zeolite treatments, and the increase of CAB Cd after nano zeolite treatment NZ3 is 1.24 times higher than that after ordinary zeolite treatment OZ3. The percentage of EXE Cd decreased, while the percentages of CAB, OM, FMO and RES Cd increased, indicating that Cd speciation that is sensitive to environmental change, has higher biological activity and toxicity and can be easily absorbed by plants gradually transformed into the speciation that is stable in the environment, has less activity and toxicity and is difficult to be absorbed. The results show that the application of nano zeolite has an advantage over ordinary zeolite in stabilizing heavy metal Cd to a certain extent and reducing their bioavailability and transferability.

4 Conclusions

Tobacco pot experiment was conducted using exogenous Cd contaminated soils. The results showed that application of ordinary zeolite and nano zeolite could increase soil pH, reduce soil available Cd content, reduce Cd contents in the upper, middle and lower leaves and stems of tobacco, promote transformation of EXE Cd in the soil to CAB, FMO, OM and RES Cd. There was a significant negative correlation between soil pH and available Cd content and significantly positive correlation between soil available Cd content and Cd content in the upper, middle and lower leaves as well as the stem of tobacco. The effect of application of nano zeolite was better than that of application of ordinary zeolite in terms of increasing pH, reducing available Cd content, decreasing Cd content in various parts of tobacco as well as reducing Cd bioavailability and transferability. Therefore, application of nano zeolite has an advantage over ordinary zeolite in situ reducing and remediating Cd contaminated soils .

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Conflict of interest: Authors state no conflict of interest.

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