Comparison of cadmium and arsenic accumulation characteristics and remediation potential of different forage plants

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Abstract: A field experiment was carried out in a farmland polluted by cadmium (Cd) and arsenic (As) in Yunnan Province to study Cd and As accumulation characteristics of 15 different forage plants, so as to screen out plants for comprehensive utilization in the farmland polluted by Cd and As. The results showed that the best growth conditions were for ‘Teosinte’ and ‘King grass’, the worst growth condition was ‘Pennisetum’. The Cd contents of shoots of ‘King grass’, ‘Sweet elephant grass’, ‘Pennisetum’, ‘Teosinte’ and ‘Alfalfa’ did not exceed the Chinese feed hygiene standard (≤ 1mg kg⁻¹). The content of total arsenic in shoot of ‘Tall fescue’ did not exceed the feed hygienic standard (≤ 4 mg kg⁻¹). ‘Sorghum12FS9003’ and ‘Sorghum×Sudan’ have the strongest ability to enrich Cd and As, which could be used for remediation of soil polluted by Cd and As. ‘Alfalfa’ has low Cd and As accumulation capacity.

1. Introduction
Heavy metal pollution will lead to a series of problems, such as the decline of crop yield and quality, and excessive heavy metals in crops, which will lead to the vicious cycle of agricultural ecosystem[1]. The over standard rates of cadmium and arsenic are 7% and 2.7% respectively based on China soil pollution survey bulletin in 2014. The two elements are widely concerned because of their wide pollution and toxic effects[2]. Phytoremediation technology has some problems, such as long remediation time and difficult to be commercialized. Therefore, it is more appropriate to select crops with different accumulation characteristics for comprehensive utilization in farmland with different pollution levels in China[3]. At present, most of the low accumulation varieties screened only aim at single heavy metal, while few varieties screened in Cd and As compound polluted soil[4]. Forage has the advantages of low planting cost, simple management, high yield, good economic benefits, and resistance under heavy metal stress[5]. In this research, fifteen kinds of different forage plants were planted in the farmland polluted by Cd and As. The contents of Cd and As in the soil, plant height, biomass, contents of Cd and As in the plant were determined, and the bioaccumulation factors and translocation factors of Cd and As in plants were analyzed in order to screen out Cd and As forage plants which meet the national feed hygiene standards or be with phytoremediation potential.
2. Materials and methods

2.1. Experimental site location and Experimental design and Indicator determination
The experimental area was located in Zhadian District, Jijie Town, north of Gejiu City, Yunnan Province, with an altitude of 1428 m, 103 ° 15 ′ E and 23 ° 46 ′ N. The soil of the experiment site was mainly contaminated by Cd and As. Physical and chemical properties were as follows: The pH between 7.64 and 8.19, 40.26 g kg⁻¹ of organic content, 2.54 g kg⁻¹ of total nitrogen content, 1.36 g kg⁻¹ of total phosphorus content, 2.26 g kg⁻¹ of total potassium content, 142.23 mg kg⁻¹ of alkali-hydrolyzable nitrogen content, 179.5 mg kg⁻¹ of available phosphorus content, 178.91 mg kg⁻¹ of available potassium, 5.09 mg kg⁻¹ Cd and 226.13 mg kg⁻¹ As, which belonged to severe pollution.

The seeds were sown in October, 2019 and randomly distributed in the field with three repetitions. Each plot area was 15 m², a total of 45 plots. ‘Sorghum’, ‘Sudan grass’, ‘Sorghum×Sudan’, ‘Sweet elephant grass’, ‘Pennisetum’, ‘Timothy’, ‘Teosinte’, ‘Chicory’, ‘Indian lettuce’ ‘Perennial ryegrass’, ‘Annual ryegrass’, ‘Alfalfa’, ‘Tall fescue’ were 30 cm in line spacing and 20 cm in sowing depth; ‘King grass’ was 50 cm in plant spacing and 60 cm in line spacing and 20 cm in planting depth. All forages were planted by artificial strips. During the growth period, insecticidal, weeding, watering and topdressing were carried out according to the growth status.

2.2. Indicator determination
Collect samples in June 2020. Plant height, root length and biomass were measured. After cleaning the plants with deionized water, they were put in an oven at 105°C for 30 min, and then dried in an oven at 75°C to a constant weight. Finally, they were crushed with a stainless steel grinder. Soil samples were ground to ≤ 0.149 mm after air drying.

The soil contents of Cd were determined by digesting with concentrated HNO₃-HClO₄, followed by flame atomic absorption spectrometry. The soil contents of As were determined by digesting with concentrated HNO₃: HCl: deionized water (3:1:4), followed by atomic fluorescence spectrometer [6].

The plants contents of Cd were determined by pressure digestion tank digesting with HNO₃: H₂O₂ (3:1, V/V), followed by flame atomic absorption spectrometry. The plants contents of As were determined by pressure digestion tank digesting with HNO₃, followed by atomic fluorescence spectrometer [7].

2.3. Statistical analyses
Bioconcentration factor (BCF) = Heavy metal content in shoot of plant (mg kg⁻¹) / Heavy metal content in soil (mg kg⁻¹)

Translocation factor (TF) = Heavy metal content in shoot of plant (mg kg⁻¹) / Heavy metal content in root of plant (mg kg⁻¹)

The data were processed by Microsoft Excel, and analyzed with SPSS 22.0. The statistical significance analyzed used one-way analysis of variance (ANOVA) and Duncan test the difference of the average value of different treatments at the 0.05 level. Figures were produced using Origin 2018.

3. Results

3.1. Difference of growth status of different forage plants under Cd and As combined stress
There were obvious differences in the growth status of different forage plants (Figure 1). The plant height of ‘Sorghum12FS9003’ was the highest, which was 180.3 cm. The root length of ‘King grass’ was the longest, which were 17.85 cm. The plant height and root length of ‘Tall fescue’ were the lowest, which was 29.56 cm and 8.5 cm, respectively. Biomass of ‘Teosinte’ was the highest, which was 5.06 kg m⁻², and that of ‘Pennisetum’ was only 0.14 kg m⁻².
3.2. Differences of Cd and As contents in shoot and root of different forage plants

As shown in Figure 2, among the 15 plants, the Cd content in the shoot of ‘Chicory’ was the highest, which reached 4.71 mg kg\(^{-1}\), and the lowest Cd content in the shoot was ‘Sweet elephant grass’, which was 0.18 mg kg\(^{-1}\). The Cd content in the root of ‘Sorghum12FS9003’ was the highest, which reached 6.78 mg kg\(^{-1}\). It had a strong Cd accumulation ability, which was 48.4 times higher than ‘Alfalfa’. According to the national feed hygiene standard of China (GB 13078-2017, Cd < 1 mg kg\(^{-1}\)), the shoot of Cd contents of 15 plants were evaluated, only ‘King grass’, ‘Sweet elephant grass’, ‘Pennisetum’, ‘Teosinte’ and ‘Alfalfa’ reached the feed hygiene standard, and the rest materials exceeded the standard in varying degrees.
Figure 3. Content of As in shoot and root of different forage plants.

The figure 3 showed that the As content in the shoot of ‘Sorghum×Sudan’ was the highest, which was 18.31 mg kg\(^{-1}\), 4.6 times higher than the feed hygienic standard (As < 4 mg kg\(^{-1}\)). Among the 15 plants, only the shoot of ‘Tall fescue’ reached the feed hygienic standard, and the As content was 3.81 mg kg\(^{-1}\). The root of ‘Perennial ryegrass’ had the strongest As accumulation ability, and the content in the root was 5.52 mg kg\(^{-1}\); the As content in the root of ‘Alfalfa’ was the lowest, which was only 4.7% of the ‘Perennial ryegrass’.

3.3. Accumulation characteristics and remediation potential of Cd and As in different forage plants

Fifteen materials had some bioconcentration capacity for Cd, with bioconcentration factor of Cd ranging from 0.13 to 0.87. The bioconcentration factor of Cd ‘Chicory’, ‘Sorghum12FS9003’ and ‘Indian lettuce’ were 0.868, 0.856 and 0.823, respectively, which were the three plants with the highest Cd bioconcentration factors among the 15 plants. ‘Sweet elephant grass’ had the weakest bioconcentration capacity for Cd, with bioconcentration factor of only 4.7% of that of the highest ‘Chicory’. The bioconcentration factor of all plants for As was less than 0.1.

The results showed that different forage plants had different transport capacity for Cd and As. The Cd translocation factor of *Leguminosae* and *Compositae* were more than 1, while those of *Gramineae* were less than 1, indicating that *Leguminosae* and *Compositae* had stronger Cd transport capacity than *Gramineae*. Most plants had a strong ability to transport As, only ‘Tall fescue’ had a translocation factor of As <1, which was 0.741. The Translocation factors of the other plants ranged from 1.249 to 9.26.

Membership function was used to evaluate the restoration potential of each material found that ‘Sorghum12FS9003’ and ‘Sorghum×Sudan’ had the phytoremediation potential of Cd and As compound pollution. The phytoremediation potential of ‘Alfalfa’ was lowest, and its ability to accumulate cadmium and arsenic from soil was low.

4. Discussions

Forage showed strong tolerance to heavy metal stress\[^8\]. The results of this study showed that the 15 tested forages plants could grow normally under the combined stress of Cd and As, indicating that these plants have a certain tolerance to the combined stress of Cd and As, which is mainly related to the plant's antioxidant system and heavy metal transporter\[^9\]and other detoxification mechanisms. The comparison of 15 tested plants showed that the plant height of *Gramineae* was generally higher than that of *Leguminosae* and *Compositae*; the biomass of ‘Sorghum12FS9003’, ‘King grass’ and ‘Teosinte’ was 2.32-5.06 kg m\(^{-2}\), and the growth status was significantly better than that of *Leguminosae* and *Compositae*, which was mainly related to the growth characteristics and interspecific differences of each material.
In terms of bioaccumulation factors, ‘Chicory’ and ‘Indian lettuce’ were more than 0.8 for Cd, and only 0.033 and 0.032 for As, respectively, indicating significant differences in enrichment ability. Cd enters plant roots by exchanging with $H^+$ on the cortical membrane of root surface, occupying non-selective cation channels (Fe$^{2+}$, Zn$^{2+}$, Ca$^{2+}$), and then transports to shoot to achieve enrichment$^{[10]}$. As (V) can enter plants through phosphorus absorption and transport channels, As (III) mainly enters plants in the form of As (OH)$^3-$ through NIPs transporters, while NIPs aquaporins can transport urea, silicic acid and other substances, resulting in a multi-competitive relationship between plants in absorbing and enriching As$^{[11]}$. These may be responsible for the differences in the enrichment of cadmium and arsenic by plants.

The results showed that the Cd translocation factors of *Gramineous* were less than 1, while those of *Leguminosae* and *Compositae* were more than 1, which indicated that Cd was more distributed in the shoot of ‘Alfalfa’, ‘Chicory’ and ‘Indian lettuce’. Daniela et al.$^{[12]}$ also found that leafy vegetables such as ‘Lettuce’, ‘Chicory’ and similar horticultural crops have high Cd absorption and transport capacity. The 15 tested forage plants in this experiment have a strong ability to transport As. The results showed that only the ‘Tall fescue’ had a translocation factor of As <1, and the rest of the tested plants were >1. The strong transport of As by the tested plants may be related to the strong transpiration and the strong reduction ability of the roots$^{[13]}$.

5. Conclusions
The Cd contents of ‘King grass’, ‘Sweet elephant grass’, ‘Pennisetum’, ‘Teosinte’ and ‘Alfalfa’ met the limit of China feed hygienic standard, and the As content of ‘Tall fescue’ met the limit of China feed hygienic standard. ‘Alfalfa’ has the potential of low accumulation ability of Cd and As. ‘Sorghum12FS9003’ and ‘Sorghum×Sudan’ have certain potential for phytoremediation of soil compound contaminated by Cd and As.

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