#### **RESEARCH ARTICLE**



# Phytoremediation with kenaf (*Hibiscus cannabinus* L.) for cadmium-contaminated paddy soil in southern China: translocation, uptake, and assessment of cultivars

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### Abstract

Kenaf (*Hibiscus cannabinus* L.) is suitable for growing in heavy metal-polluted soil for non-food purposes and can be used as a potential crop to remediate heavy metal-contaminated soil. The main objective of this study was to investigate kenaf phytoextraction of cadmium (Cd), including uptake, translocation, and accumulation differences in tissues among kenaf cultivars. A field experiment was conducted in a Cd contaminated paddy field in southern China area with 13 kenaf cultivars in 2015 and 2016. Agronomic performance, Cd concentrations in plant tissues (root, xylem, and phloem), and biomass of different tissues of each cultivar were measured and evaluated. Significant differences in Cd concentrations and accumulation among tissues and cultivars were observed. The phloem had the highest Cd accumulation and transfer capability compared with the roots and xylem. Approximately 35 ~ 65 g of Cd could be taken up by the aerial parts of different kenaf cultivars within every hectare of soil. The percentage of Cd uptake by the phloem ranged from 47 to 61% and by the xylem ranged from 38 to 53%. By evaluating the agronomic traits and Cd bioaccumulation capacity, Fuhong 952, Fuhong 992, and Fuhong R1 were regarded as Cd accumulators for the phytoremediation of Cd-contaminated soil. Our study clearly demonstrated that a significant level of Cd in the soil was taken up through the phytoremediation with kenaf. In addition, harmless utilization of kenaf planting in Cd-contaminated paddy soil was discussed.

Keywords Heavy metal · Kenaf · Soil pollution · Phytoextraction · Phloem · Bioconcentration factor · Translocation factor

# Introduction

In recent decades, the rapid industrialization and economic development have not only improved people's living standards but also induced serious contamination to the farmland in China. There are about 19.4% of the agricultural lands suffering from heavy metals and metalloid pollution according to the latest nationwide surveys. Among the metals or metalloid, cadmium (Cd) and arsenic (As) are the

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main contaminations to soils in China (Chen et al. 2020). Although Cd is non-necessary and even harmful to crops, it can still be easily assimilated by the plant root and consequently transported to their tissues. It was reported that rice plant could take in Cd from the soil and accumulate it with a high level in its aerial tissues even if the Cd content in soil was very low (Wang et al. 2011). The Chinese limit of Cd in brown rice is  $0.2 \text{ mg kg}^{-1}$  (GB 2762–2012). The CODEX commission of FAO/WHO stipulates that the Cd content in the cereal grains rice and wheat should be < 0.4 and < 0.2, respectively. Hence, restoring the heavy metal-contaminated paddy lands becomes an urgent task for China. As an in situ and nondestructive remediation approach, phytoremediation cannot only remediate the polluted soil but produce feedstock for further use. Apart from common heavy metal hyperaccumulators, some crops have also been used for phytoremediation, such as alfalfa, tobacco, flax, jute (Liu et al. 2019; Zhang et al. 2019; Guo et al. 2019, 2020). Since the removal rate of heavy metal depends on both the plant biomass and the metal content involved in it, the selection of a suitable plant becomes a key point in phytoremediation (Sumiahadi and Acar 2018; Guo et al. 2020).

Kenaf (Hibiscus cannabinus L.) is one of the most important fiber crops with high economic benefits, and massive heavy metals accumulated found in kenaf do not enter the food chain (Deng et al. 2017; Chen et al. 2021). Kenaf plants grow rapidly with a large biomass, and the plants can grow to a height of 4-6 m and yield fresh biomass of up to 100-150 t per hectare within a 4-month growth period (Tao et al. 2007). Previous studies showed that kenaf is a potential plant for use in the phytoremediation of widespread heavy metal contamination. Kenaf plants can remove 89.2% of Cd in contaminated water by rhizofiltration (Niazy 2015). During 3 months of growth in sludge, kenaf accumulated 2.49 mg kg<sup>-1</sup> Cd and 82.5 mg kg<sup>-1</sup> zinc without obvious toxic symptoms (Arbaoui et al. 2013). However, most of the research results were based on pot experiments, which are generally incompatible with large-scale phytomanagement owing to the interactions of many other factors affecting plants in the agricultural land. The actual Cd absorb ability of kenaf in the field was not clear, and the characteristics of Cd accumulation in phloem, the most economically part of kenaf, have not been studied.

In the present study, the responses and phytoremediation potential of 13 different kenaf cultivars to Cd contamination were investigated in a field trial. In detail, the main objectives of this study were to (i) compare the differences of Cd translocation and accumulation in kenaf tissues between cultivars; (ii) analyze the bioconcentration factors and translocation factors for Cd of kenaf cultivars; and (iii) evaluate the phytoremediation potentials of kenaf cultivars and find more suitable cultivars for phytoremediation.

### **Material and methods**

### **Experimental site and soil**

The field experiment was conducted at the garden village of Zhuzhou city, Hunan province, China (27°42'58.52"N, 113°11'8.26"E). The composition of soil particles is 52% clay, 22% silt, and 26% sand. The cation exchange capacity (CEC) is 11.2 cmol kg<sup>-1</sup> (+). The content of soil organic matter, available phosphorus, and available potassium are 31 g kg<sup>-1</sup>, 1.42 mg kg<sup>-1</sup>, and 79.33 mg kg<sup>-1</sup>, respectively. The plumbum (Pb), Cd, and As contents of soil were determined, and the result showed that the average Cd content was 2.25 times higher than that of the standard of Chinese paddy soil (Table 1). According to the standard for soil environmental quality and risk control of soil pollution in agricultural land, the soil of the experimental field was Table 1 Basic properties of the tested paddy soil

Items	рН	Pb (mg/kg)	Cd (mg/kg)	As (mg/kg)
Range	4.8~5.7	70.4~118.1	0.373~1.443	9.5~12.8
Average	5.2	98.0	0.9	11.5
Standard (GB15618- 2018)	5.5~6.5	100	0.4	30

classified as moderately Cd-contaminated agricultural soil (GB 15,618–2018).

#### Field experiment and sample collection

Thirteen kenaf cultivars were sown in the field in May 2015 and 2016. Among these cultivars, DGG, G4, GGMN, and GGS were not Chinese native varieties, with unlobed leaf shapes. The other nine cultivars were Chinese varieties that are mainly cultivated in the south of the Yangtze River. The leaves of these nine cultivars were partially lobed or deeply lobed (Fig. 1). There were total 39 plots (13 cultivars  $\times 3$ replications) for growing kenaf in this experiment. Each plot was 2 m wide and 3 m long, and all of them were arranged with a random blocked design. The seeding quantity of each plot was based on the seedling rate of cultivars and 40 seeds per square meter. The kenaf was harvested in mid-November of both 2015 and 2016; 10 kenaf plants which uniformly distributed in each experimental plot were collected and mixed to obtain a sample. Then, the aerial parts of plants in each plot were harvested to obtain the fresh weight and dry weight. The leaves of kenaf were not considered as phytoextraction tissue in this study since most of the leaves have fallen when harvested.

The samples were taken to the laboratory immediately, and plant height, stem diameter, and skin thickness were measured for each plant. The middle part of the stem is selected for measuring the stem diameter and skin thickness with vernier caliper. Then, the root, xylem, and phloem were separated by hand. After shaking off the rhizosphere soil in a plastic bag, the roots are separated with a guillotine, washed with tap water until the surface is free of soil and impurities, then washed repeatedly with deionized water for  $3 \sim 5$  times, and the water on the sample surface is sucked dry with absorbent paper. The phloem (bark) was peeled manually, and the xylem (stick) of the peeled stem is collected. The samples were collected and put into paper bags respectively. After weighting the fresh weight, the samples were placed in a drying oven 105 °C for 30 min, then 70 °C until the weight of the samples become constant, and grounded with a stainless-steel grinder (1000Y, China). The rhizosphere soils eliminated the mixed tiny root, and were air dried and sieved through a 60-mesh sieve for testing.

Fig. 1 Different leaf shapes of 13 kenaf cultivars



### **Chemical analysis**

The testing of CEC, soil particle composition, organic matter, and soil nutrient followed the methods described in the agricultural industry standard of the People's Republic of China (NYT1121.3: Soil testing). The chemical parameters of soil and plant samples were analyzed following the methods described in our previous study (Guo et al. 2020). The pH of soil samples were measured in a 1:2.5 soil-towater suspension using a glass electrode (PHS-25, Leici, China). The soil samples were fully dissolved with hydrochloric-nitric-hydrofluoric-perchloric acid; the concentrations of Cd and Pb were determined by atomic absorption spectroscopy method (Z2310, Hitachi, Japan). For As, the soil samples were acid digested with aqua regia, and the concentration was then determined using the atomic fluorescence method (AFS-8230, Titan, China). The plant samples were digested with nitric-perchloric acid, and then the digestion solution was used to determine the metal concentration with a graphite furnace atomic absorption spectrometry (Z2000, Hitachi, Japan).

### **Bioconcentration factor and translocation factor**

The bioconcentration factor (BF) and translocation factor (TF) were calculated to evaluate the accumulation and translocation of heavy metals of different kenaf cultivars (Yoon et al. 2006). BF is the ratio of metal concentration in the root to rhizosphere soil. TF is the ratio of metal concentration in the xylem to root (TFrx) and phloem to root (TFrp). BF and TF can be used to estimate accumulation and translocation capability of plant and the redistribution of metals throughout plant tissues.

#### **Statistical analysis**

The differences among the kenaf cultivars were evaluated using one-way analysis of variances and Duncan's multiple range test. All statistical analyses were performed using the statistics package Excel 2010 and SPSS v19.0.

### Results

# Agronomic traits and fresh weight of kenaf cultivars in Cd-contaminated paddy soil

The heights of the kenaf plants were between 2.5 and 4.0 m in the two growing seasons. Most of cultivars grew higher in 2016 than in 2015, except for Hongma No. 1 and Fuhong 952. The average plant height in the growing season in 2015 and 2016 was 3.1 and 3.3 m, respectively. The stem diameter of kenaf plants was between 13.0 and 19.1 mm in the two growing seasons. All of cultivars had a higher diameter in 2016 than in 2015. The average diameter in the growing season in 2015 and 2016 was 15.0 and 17.0 mm, respectively. The bark thickness of the kenaf plants was between 0.84 and 1.48 mm in the two growing seasons. Most of cultivars had thicker bark in 2016 than in 2015, except Hongma No. 1 and Fuhong 952. The average bark thickness during the growing season in 2015 and 2016 was 1.02 and 1.22 mm, respectively. The fresh weights the of root, xylem, and phloem for 10 plants were respectively between 0.18 and 0.32 kg, 0.94 and 2.40 kg, and 0.62 and 1.60 kg, respectively. Among the 13 cultivars, the fresh weights of the root, xylem, and phloem for Fuhong 952 were more stable in the two seasons and relatively higher than those of the other cultivars (Fig. 2).



Fig. 2 Agronomic traits and fresh weight of kenaf cultivars in Cd-contaminated paddy soil





# Cd concentration in rhizosphere soil and different tissues of kenaf plants

The Cd concentration in the rhizosphere soil in 39 experimental pots was between 0.71 mg kg<sup>-1</sup> and 1.50 mg kg<sup>-1</sup>, with a mean concentration of 1.15 mg kg<sup>-1</sup> (Fig. 3). The Cd concentrations in the rhizosphere soil of Hongzong No. 1, Hongyou No. 2, Fuhong 952, Fuhong 992, and Fuhong R1 were significantly higher than those of the other cultivars. The Cd concentrations in kenaf tissues are presented in Table 2. The Cd concentration ranges in the root, xylem, and phloem of 13 kenaf cultivars were 1.50–4.38 mg kg<sup>-1</sup>, 1.00–2.96 mg kg<sup>-1</sup>, and 2.39–5.18 mg kg<sup>-1</sup>, respectively, with mean concentrations of 2.67, 2.05, and 3.87 mg kg<sup>-1</sup>, respectively. The mean accumulation of Cd in the three

tissues of kenaf was decreased in the following order: phloem  $^{\circ}$  root  $^{\circ}$  xylem, and the mean concentrations of Cd in the phloem of the 13 cultivars were 1.1–2.9 times higher than those in the root and xylem. The top three kenaf cultivars with the highest Cd concentrations were Hongzong No. 1, Fuhong 992, and Fuhong 952.

# Bioconcentration and translocation of Cd in kenaf tissues

BF was used to evaluate the ability of the kenaf plant to accumulate Cd in roots of the 13 kenaf cultivars. BFs of roots of the 13 kenaf cultivars ranged from 1.74 to 2.96, with a mean of 2.35. No significant difference was found among cultivars, except for Fuhong 992 and GGMN, which Table 2 Cd concentrations in kenaf tissues with 13 kenaf cultivars

Cultivars	Tissues	Tissues			Average
	Root	Xylem	Phloem		
Hongma No. 1	$2.32 \pm 0.28$ ef	1.26±0.27 f	2.54±0.15 e	1.00~2.69	2.04
Hongma No. 2	$2.52 \pm 0.19$ e	$1.63 \pm 0.28$ def	2.89±0.17 e	1.36~3.05	2.35
Hongma No. 3	$2.36 \pm 0.32$ e	$1.43 \pm 0.25$ ef	2.69±0.28 e	1.20~2.99	2.16
Hongma No. 5	1.91±0.16 gh	$1.5 \pm 0.21$ ef	2.73±0.23 e	1.29~2.96	2.05
Hongzong No. 1	$3.74 \pm 0.17$ ab	$2.79 \pm 0.22$ a	4.75±0.11 ab	2.53~4.86	3.76
Hongyou No. 2	$2.93 \pm 0.2 \text{ d}$	$2.05 \pm 0.16$ bcd	$4.69 \pm 0.24$ abc	1.87~4.88	3.22
Fuhong 952	$3.31 \pm 0.22$ c	$2.57 \pm 0.3$ a	$4.33 \pm 0.35$ bc	2.32~4.73	3.40
Fuhong 992	$4.09 \pm 0.25$ a	$2.05 \pm 0.23$ bcd	$4.23 \pm 0.27$ c	1.80~4.51	3.46
Fuhong R1	$3.43 \pm 0.09$ bc	$2.75 \pm 0.17$ a	$3.5 \pm 0.16$ d	2.62~3.64	3.23
DGG	$2.16 \pm 0.19$ fg	$2.42 \pm 0.25$ ab	$4.33 \pm 0.24$ bc	1.94~4.54	2.97
G4	$1.95 \pm 0.19$ gh	$2.14 \pm 0.25$ bc	$4.43 \pm 0.33$ bc	1.75~4.67	2.84
GGMN	$1.66 \pm 0.2$ h	$2.35 \pm 0.23$ ab	$4.26 \pm 0.42$ c	1.50~4.71	2.76
GGS	2.37±0.23 e	$1.73 \pm 0.18$ cde	4.94±0.22 a	1.63~5.68	3.01
Range	1.50~4.38	1.00~2.96	2.39~5.18	_	_
Average	2.67	2.05	3.87	_	_

The different lowercase letters in a column indicate significant differences among tissues at P < 0.05



Fig. 4 Bioconcentration factor (BF) and translocation factor (TF) of Cd in kenaf tissues. The different lowercase letters indicate significant differences among cultivars at P < 0.05

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showed the highest and lowest BFs in the 13 kenaf cultivars, respectively (Fig. 4). TF was used to evaluate the ability of a kenaf plant to transfer Cd within its tissues. TFrxs and TFrps of kenaf cultivars ranged from 0.50 to 1.44 and 1.10 to 2.57, respectively (Fig. 4). TFrps were higher than TFrxs in all 13 cultivars, suggesting the phloem had a greater ability to transport Cd that of the xylem. There were significantly higher differences among the TFrps of the cultivar than among the TFrx and BF of the roots, indicating a difference in the Cd transport capacity of the phloem among kenaf varieties. TFrps of cultivars with an unlobed leaf shape (DGG, G4, GGMN, and GGS) were significantly higher than those of cultivars with other leaf shapes (Fig. 4).

### Biomass of kenaf planting in Cd-contaminated paddy soil and assessment of Cd uptake for the 13 kenaf cultivars

The biomass of the root, xylem, and phloem of 10 plants among the 13 cultivars were 0.06–0.10 kg, 0.17–0.36 kg, and 0.13–0.36 kg, respectively. The biomass of the xylem was higher than that of other tissues in all 13 kenaf cultivars, accounting for 46–55% of the total biomass. Phloem biomass was lower than xylem biomass but higher than root biomass. The percentage of phloem biomass ranged from 29 to 38%, and the root biomass ranged from 14 to 19%. Fuhong 952 (0.72 kg) and G4 (0.37 kg) had the highest and lowest of total 10 plants biomass (Fig. 5).

In the actual agricultural operations, only the aerial parts of kenaf plants are harvested. According to the data of fresh weight and dry weight by experimental plots, and the Cd concentration in different tissues, we assessed Cd uptake of each cultivar for a hectare area. Because all leaves had fallen off during harvest, the biomass of leaves was not calculated in the total aerial biomass. Fuhong 952 and Fuhong R1 had the highest Cd phytoextraction capacity and uptake exceeding 60 g of Cd from every hectare of soil. For most cultivars, except for Fuhong R1, phloem phytoextraction accumulated more Cd than xylem. The percentage of Cd uptake by the phloem ranged from 47 to 61%, and that by the xylem ranged from 38 to 53% (Fig. 6).

# Comprehensive evaluation of the 13 kenaf cultivars in the phytoremediation of Cd-contaminated soil

To comprehensive evaluate the 13 kenaf cultivars in the phytoremediation of Cd-contaminated soil, we normalized and clustered the data using the zero-to-one method in the heat map (Fig. 7). Fuhong 992, Fuhong 952, and Fuhong R1 exhibited better performance than other cultivars in most of the traits, except for the TFs of the xylem and phloem, and thus, they can be used as candidate plant materials for soil phytoremediation.

### Discussion

### Differences in Cd accumulation and phytoextraction potential of kenaf cultivars for Cd-contaminated soil phytoremediation

In the present study, differences have been detected in the bioaccumulation of Cd in different plant tissues; the result is the same as previously reported (Li et al. 2018). Differences



Fig. 5 Biomass of kenaf cultivars planting in Cd-contaminated paddy soil

Fig. 6 Assessment of Cd uptake

for the 13 kenaf cultivars





Fig. 7 Comprehensive evaluation of the 13 kenaf cultivars in the phytoremediation of Cd-contaminated soil

were also detected in Cd uptake among the kenaf cultivars in the present study. However, differences were not detected in Cd concentrations of different kenaf cultivars when soil was severely contaminated with other heavy metals, such as Zn, Cu, and Cr (Li et al. 2018). It is speculated that the interaction between metal elements may affect the absorption of Cd in kenaf plants (Chen et al. 2016).

The total Cd uptaking from the square unit of the soil is an important index to evaluation the ability of kenaf for phytoremediation. In this study, the Cd uptake of 13 kenaf cultivars planted in 1 ha of soil was between 39.8 and 64.5 g. In our experiment, we used a single heavy metal-contaminated soil. In compound heavy metal-contaminated soil (Cd, Zn, Ni, Cu), 25.7 g ha<sup>-1</sup> of Cd can be phytoextracted by kenaf plants (Li et al. 2018), which is less than that by Hongma No. 5, which takes up the least amount of Cd in our study, possibly because other heavy metals affect the absorption of Cd. Although the roots of kenaf immobilized large portions of the total plant Pb from the sand tailings (Mo et al. 2008), in the present study, root biomass only accounted for 14–19% of the total plant, indicating that it is not the main Cd phytoextraction tissue of Cd in kenaf plants. In addition, root harvesting is not practical for large-scale farmland cultivation of kenaf.

### Cd accumulation in the phloem of kenaf plant

Previous studies have reported heavy metal bioaccumulation in the roots, stems, and leaves of kenaf plants (Arbaoui et al. 2013; Li et al. 2018). However, these studies did not consider Cd accumulation in the phloem, which is an economically and important part of kenaf, and impact the safety of subsequent utilization, such as fiber from phloem. In the present study, Cd concentration of the phloem ranged from 2.39 to 4.94 mg kg<sup>-1</sup>, with the Cd concentration of soil being  $0.373-1.443 \text{ mg kg}^{-1}$ , proving that phloem accumulated the highest amount of Cd in the soil. The mean concentrations of Cd in the phloem of 13 cultivars were 1-3 times higher than those in other tissues. This indicates that the phloem is the greatest accumulator tissues of kenaf, rather than the root. The same results were previously verified in flax and jute plants, which are bast fiber crops such as kenaf (Guo et al. 2019, 2020). We recently found that Cd concentration in raw ramie fiber is 1.5–8.5 times that in its leaf and xylem, which varies according to ramie varieties (Wu et al. 2021). Within the shoot of rice, phloem transport is the basis for Cd redistribution and for the accumulation in fruits and seeds (Satoru et al. 2012; Page and Feller 2015). In potato plants,

Cd can be rapidly distributed to all tissues via the phloem (Reid et al. 2010). This was also observed in hyperaccumulator (Lu et al. 2013; Wei et al. 2014; Feller et al. 2019). In this study, we found that the differences in leaf shapes of kenaf cultivars lead to the different Cd translocation capability of phloem. It provides a clear basis for variety selection in the followed phytoremediation work. However, in-deep studies still needed to obtain more details on the distribution and accumulation of Cd in the phloem of bast fiber crops.

# Harmless utilization of kenaf planting in Cd-contaminated paddy soil

The Cd uptake of hyperaccumulator Sedum alfredii planting in mildly  $(0.53 \text{ mg kg}^{-1})$  and moderately  $(1.55 \text{ mg kg}^{-1}) \text{ Cd}$ contaminated soil was 40 g ha<sup>-1</sup> and 241 g ha<sup>-1</sup>, respectively (Zhu et al. 2019). Amaranthus hypochondriacus L. could extract  $13 \sim 39$  g ha<sup>-1</sup> of Cd when the Cd concentration of soil was 0.71 mg kg<sup>-1</sup> (Song et al. 2019). The phytoremediation capacity of kenaf is not similar to that of Sedum alfredii but better than that of Amaranthus hypochondriacus L. Compared with these hyperaccumulator plants, large biomass is an advantage of kenaf; in our study, maximum 25 t of dry biomass can be obtained in a hectare of soil. For the harvested kenaf straw enriched with Cd, it is important to reuse it not only to limit secondary environmental pollution but also to provide both economic and social benefits. The authors believe that the application of kenaf straw as a building material for the phytoremediation of Cd-contaminated soil can meet the above requirements. Kenaf fibers have good mechanical properties, the average tensile strength of kenaf fiber is between 157 and 600 MPa, and the average ultimate tensile strain and elastic modulus are 0.015-0.019 and 12,800-34,200 MPa, respectively (Symington et al. 2009; Shahar et al. 2019). The use of kenaf fiber to replace traditional rayon and synthetic fibers to produce fiber cement-based materials has great advantages, such as being 100% biodegradable, environmentally friendly, and low cost (Elsaid et al. 2011; Mahzabin et al. 2018). Cd enriched in kenaf straw does not affect the strength of the cement mortar, and the leaching concentration of the heavy metal is far below the prescribed safety limit (Liu et al. 2020).

# Conclusions

In the present study, 13 kenaf cultivars were cultured on moderately Cd-contaminated agricultural soil, and there were significant differences that existed in Cd accumulation among plant tissues, as well as among the cultivars. In all kenaf cultivars, phloem accumulated more amount of Cd than the roots and xylems. The Cd uptake of some kenaf cultivars, such as Fuhong 952 and Fuhong R1, was more than  $60 \text{ g ha}^{-1}$  per season. Fuhong 992 has excellent agronomic performance compared with that of other cultivars. These kenaf cultivars can be used as candidate plant materials for soil phytoremediation. Planting these high Cd-accumulating cultivars can gradually decrease the Cd concentration of soil to natural levels and make it possible to plant edible crops again. Moreover, the aerial parts of kenaf that accumulated Cd in dry matter can be safely reused as industrial raw materials in the future.

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Author contribution YG performed the experiments, writing—original draft, and provided financial supporting. QX, XZ, and ZW participated in the sample collection. ZD, MZ, CQ, and SL analyzed most of the data; YW provided the materials and financial support. All the authors reviewed the draft and approved the final manuscript.

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**Data availability** The availability of data and materials during the current study is available from the corresponding author on reasonable request.

#### Declarations

Ethics approval and consent to participate This study has not directly involved humans or animals. No specific permission was required for use of these materials for experimental purposes. The plants were grown in the experimental field of Institute of Bast Fiber Crops, China as per standard practices, and samples were harvested at the required time. We comply with the Convention on the Trade in Endangered Species of Wild Fauna and Flora.

**Consent for publication** The manuscript was reviewed, and all authors consented to publish.

Competing interests The authors declare no competing interests.

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