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*Research Article*

# Potential of *Igniscum sachalinensis* L. and *Salix viminalis* L. for the Phytoremediation of Copper-Contaminated Soils

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The potential of *Salix viminalis* L. and *Igniscum sachalinensis* L. for phytoremediation of copper- (Cu-) contaminated soils was studied under greenhouse conditions. Approximately 5 kg of potted agricultural and sewage amended soils sampled from the top 0 to 20 cm depth in Neuruppin, Germany, was treated with $CuSO_4$ at concentrations 0 (control), 250, 750, and 1250 mg Cu kg$^{-1}$ soil and ethylenediaminetetraacetic acid (EDTA) at 1000 mg kg$^{-1}$ soil, respectively. Each plant species was grown on four replicates of each soil treatment. Copper accumulated in aboveground tissues tends to increase with increasing soil Cu concentration and was the lowest in stem and leaf of both plant species grown on control soils. At 750 and 1250 mg Cu kg$^{-1}$ soil, Cu accumulated in stem and leaf of *I. sachalinensis* increased by over 12- and 20-fold, respectively, whereas there was no vegetative growth in *S. viminalis* beyond 250 mg Cu kg$^{-1}$ soil. Application of EDTA to sewage amended soils increased Cu accumulated in the stem and leaf, especially in *I. sachalinensis*. In general, *I. sachalinensis* seems to have the potential to tolerate high soil Cu content and simultaneously bioaccumulate Cu in tissues and thus may have better prospects for phytoremediation.

## 1. Introduction

Globally, soil resources are increasingly being exposed to heavy metal contamination through mining and agricultural activities, waste disposal, dust deposition from foundries and smelters, industrial accidents, and direct application of fungicides and sewage sludge [1]. Although some heavy metals, including Cu, are essential for metabolic activities when present in trace amounts, they tend to be toxic at concentrations exceeding threshold limits [2]. Large soil Cu content may have adverse effect on plant growth and persist in the food chain at levels toxic to animals. With reference to the German Law for soil protection [3] Cu concentrations between 2 and 40 mg kg$^{-1}$ of total Cu and beyond 1 mg kg$^{-1}$ available Cu are critical for soil quality.

In recent times, phytoremediation has received much attention as a feasible option for extracting heavy metals from moderately polluted soils [4] compared to other *in situ* and *ex situ* remedial approaches [5]. Besides, it is economically viable [5] and environmentally sound practice [4]. Although several conditions must be met for effective phytoremediation, bioavailability of the target metal is an important requirement [6]. However, in some cases, Cu uptake by plants is usually low due to complex formation with organic matter and adsorption by oxides and clay mineral lattices, which limits its availability for plant uptake [6]. Thus, to enhance Cu availability for plant uptake, several chelating agents including ethylenediaminetetraacetic acid (EDTA), ethylenediamine disuccinic acid (EDDS), and diethylenetriamine pentaacetic acid (DTPA) [7, 8] have been investigated. In this study, EDTA was used since it remains one of the most successfully and widely used chelators with the ability to form relatively stable metal complexes in soils [9]. However, metal-EDTA complexes tend to be nonreadily biodegradable and may persist in the soil environment in the long run [10–12].

Several plant species are known to preferentially phytoextract heavy metals from soils and metabolically convert them into inactive complexes [13]. Such plant species are usually hyperaccumulators adapted to translocation and bioaccumulation of large amounts of heavy metals in shoots and roots [14, 15].

The use of plants of the genus *Salix* for phytoremediation of metal-contaminated soils has received much attention in recent times [16]. Different species of *Salix* as well as some clones vary considerably with respect to metal translocation patterns and tolerance to heavy metals [17]. Tolerance to heavy metals such as Cd, Cu, and Zn has been documented for few European *Salix* spp. [18], and several studies have focused on their potential for phytoextraction of heavy metals from soils [5, 16, 19–21]. Furthermore, plants of the genus *Igniscum* are known to grow under adverse ecological conditions with the potential to accumulate heavy metals [22]. However, review of the literature suggests that *Igniscum* spp. have rarely been explored for phytoremediation.

Both *Salix viminalis* L. and *Igniscum sachalinensis* L. are generally cultivated in the study area for bioenergetic purpose. However, their potential for phytoremediation of Cu-contaminated soils has not been extensively studied. The objective of this study was therefore to assess the potential of *S. viminalis* and *I. sachalinensis* to tolerate and phytoextract Cu from agricultural soils treated with increasing Cu concentrations and from sewage amended soils treated with EDTA with focus on Cu accumulation in stem and leaf.

## 2. Material and Methods

*2.1. Soil.* Soil samples were collected at the top 0 to 20 cm depth from agricultural and long-term sewage amended fields in Neuruppin close to Berlin, Germany. Both fields were not under cultivation as at the time of sampling. At each site, multiple core soil samples were randomly collected with an auger. Soils sampled at each site were thoroughly mixed and bulked together to form a composite of about 250 kg. The samples were freed of plant materials and stones before storage in plastic bags and subsequently transported to the greenhouse. At the greenhouse, soil samples were air-dried, homogenized, and passed through 2 mm mesh after which portions were taken for chemical analysis. Some chemical properties of the agricultural and sewage amended soils are presented in Table 1.

*2.2. Plant Species.* *Salix viminalis* L. belongs to the Salicaceae family and it is noted to tolerate a wide range of soil types. Besides, it can be easily propagated, is fast growing, and produces high biomass with the ability to resprout after harvesting of aboveground biomass. *Igniscum sachalinensis* L. is a new and distinct cultivar, belonging to the Polygonaceae family. It is also a fast growing perennial herbaceous plant characterized by a high biomass yield and can thrive on a wide range of soils (see www.conpower.de). The characteristics of both plant species make them attractive for phytoremediation.

*2.3. Greenhouse Experiment.* The experiment was set up in a complete randomized design with four replicates for each soil treatment. About 5 kg of air-dried agricultural and long-term field sewage amended soils were weighed into greenhouse pots. The potted agricultural soils were treated with Cu salt ($CuSO_4$) to obtain soil Cu contents of 0 (control), 250, 750, and 1250 mg Cu $kg^{-1}$ soil. The potted sewage amended soils were treated with EDTA (1000 mg $kg^{-1}$ soil) and without EDTA. Treated soil samples were then thoroughly mixed mechanically to ensure homogeneity. *Salix viminalis* (from sprouted stem cuttings) and *I. sachalinensis* (from seedling) were transferred and grown on the potted soils with each plant species having four replicates for each soil treatment. The experimental setup was kept under controlled conditions in the greenhouse. Ambient temperature fluctuated between 20°C and 27°C and average relative humidity was around 80% with natural photoperiod. Soil water content was examined on regular basis and deionised water added when necessary to ensure adequate soil water content throughout the experiment.

After 60 days of vegetative growth, aerial tissues of *S. viminalis* (without the original cuttings) and *I. sachalinensis* were harvested, separated into stem and leaf, and oven-dried at 45°C to a constant weight. The stem and leaf were further oven-dried at 70°C for 48 hours, finely ground into powdery form using a nonmetal grinder, and then stored in glass bottles for subsequent analysis of Cu content. Approximately 100 g soil was taken from each pot after harvesting of the aboveground biomass of plant species. The soils were air-dried at room temperature, finely pulverized, and stored in glass bottles for subsequent laboratory analysis.

*2.4. Analytical Procedure.* Soil pH was measured in 1 : 2.5 (weight/volume basis) soils to 1 M $CaCl_2$ suspension using glass electrode. Soil organic carbon ($C_{org}$) and total nitrogen (NT) contents were determined using CNS elemental analyzer (LECO CNS 1000). Soil total Cu ($Cu_{\text{-TOT}}$) was extracted with aqua regia (1 part of concentrated $HNO_3$ with 3 parts concentrated HCl). For the determination of soil available Cu ($Cu_{\text{-PAV}}$), 20 g air-dried soil (<2 mm) was weighed into 100 mL plastic bottle, and 50 mL of 1 M $NH_4NO_3$ solution was then added, thoroughly stirred, and mechanically shaken for 2 hours after which the sample was allowed to settle for about 15 minutes before centrifugation and filtration through a 45 $\mu$m filter membrane. The filtrate was preserved by adding 0.5 mL of 1% $HNO_3$. For each batch of extraction, a blind sample was included as control. Both $Cu_{\text{-TOT}}$ and $Cu_{\text{-PAV}}$ were determined using Inductively Coupled Plasma Mass Spectrometer (ELAN 6000 ICP-MS, Perkin-Elmer Corporation). For the determination of accumulated Cu contents in stem ($Cu_{\text{-STEM}}$) and leaf ($Cu_{\text{-LEAF}}$) of *S. viminalis* and *I. sachalinensis*, approximately 0.02 g aliquot of the finely ground plant tissue was digested in 5 mL of $HNO_3$ and 1 mL of 1% hydrofluoric acid (HF) in a Teflon beaker followed by heating in an oven for 12 hours until all plant material was completely digested. The digests were filtered through 45 $\mu$m filter membrane into 50 mL plastic bottle and brought to volume by adding deionised water. Copper content in stem and leaf was also determined on the Inductively Coupled Plasma Optical Emission Spectrometer (ELAN 6000 ICP-OES, Perkin-Elmer Corporation).

*2.5. Statistical Analysis.* Statistical software, Sigma Plot (Version 12) for Windows, was used for data analysis. Two-way

Table 1: Initial pH, organic carbon ($C_{org}$), total nitrogen (TN), total phosphorus (TP), and Cu and Zn contents of the agricultural and sewage amended soils used in the greenhouse experiment.

| Soil type | pH [$CaCl_2$] | $C_{org}$ [mg $g^{-1}$ soil] | TN [mg $g^{-1}$ soil] | TP [mg $g^{-1}$ soil] | Cu [mg $kg^{-1}$ soil] | Zn [mg $kg^{-1}$ soil] |
|---|---|---|---|---|---|---|
| Agricultural soils | 7.1 | 8.8 | 1.5 | ND | 18.8 | ND |
| Sewage amended soils | 5.3 | 8.3 | 1.6 | 4.7 | 687.0 | 100.0 |

ND: not determined (concentration below the detectable limit, i.e., >0.2 mg $kg^{-1}$).

Table 2: Soil pH, total soil Cu ($Cu_{\text{-TOT}}$), available soil Cu ($Cu_{\text{-PAV}}$), and accumulated Cu in harvested stem ($Cu_{\text{-STEM}}$) and leaf ($Cu_{\text{-LEAF}}$) of *S. viminalis* and *I. sachalinensis* grown on agricultural soils treated with different levels of Cu concentration.

| | pH [$CaCl_2$] | $Cu_{\text{-TOT}}$ [mg $kg^{-1}$ soil] | $Cu_{\text{-PAV}}$ [mg $kg^{-1}$ soil] | $Cu_{\text{-STEM}}$ [mg $kg^{-1}$ DM] | $Cu_{\text{-LEAF}}$ [mg $kg^{-1}$ DM] |
|---|---|---|---|---|---|
| 0 mg Cu $kg^{-1}$ soil | | | | | |
| *S. viminalis* | $6.9 \pm 0.0^{a}$ | $22 \pm 6^{a}$ | $0.1 \pm 0.0^{a}$ | $11 \pm 3^{a}$ | $13 \pm 1^{a}$ |
| *I. sachalinensis* | $6.9 \pm 0.2^{a}$ | $16 \pm 2^{a}$ | $0.1 \pm 0.0^{a}$ | $8 \pm 3^{a}$ | $9 \pm 4^{a}$ |
| 250 mg $kg^{-1}$ soil | | | | | |
| *S. viminalis* | $6.4 \pm 0.8^{a}$ | $225 \pm 43^{a}$ | $3.5 \pm 1.8^{a}$ | $15 \pm 3^{a}$ | $20 \pm 3^{a}$ |
| *I. sachalinensis* | $6.6 \pm 0.1^{a}$ | $244 \pm 29^{a}$ | $4.7 \pm 2.6^{a}$ | $11 \pm 4^{a}$ | $147 \pm 61^{b}$ |
| 750 mg Cu $kg^{-1}$ soil | | | | | |
| *S. viminalis* | $5.9 \pm 0.9^{a}$ | $635 \pm 174^{a}$ | $64.3 \pm 39.7^{a}$ | NVG | NVG |
| *I. sachalinensis* | $5.8 \pm 0.1^{a}$ | $637 \pm 60^{a}$ | $15.8 \pm 3.7^{b}$ | 162 ± 171 | 274 ± 271 |
| 1250 mg Cu $kg^{-1}$ soil | | | | | |
| *S. viminalis* | $5.3 \pm 1^{a}$ | $1236 \pm 60^{a}$ | $117.3 \pm 50.6^{a}$ | NVG | NVG |
| *I. sachalinensis* | $5.2 \pm 1^{a}$ | $1163 \pm 162^{a}$ | $123.2 \pm 27.2^{a}$ | 299 ± 269 | 112 ± 73 |
| Cu treatment [mg $kg^{-1}$] | | | | | |
| 0 | $6.9 \pm 0.2^{a}$ | $19 \pm 5^{a}$ | $0.1 \pm 0.0^{a}$ | $9 \pm 3^{a}$ | $11 \pm 3^{a}$ |
| 250 | $6.5 \pm 0.1^{a}$ | $235 \pm 4^{b}$ | $4.1 \pm 2.2^{a}$ | $13 \pm 4^{a}$ | $84 \pm 79^{b}$ |
| 750 | $5.8 \pm 0.6^{b}$ | $636 \pm 1^{c}$ | $40.1 \pm 36.8^{b}$ | $162 \pm 171^{*}$ | $274 \pm 271^{*}$ |
| 1250 | $5.3 \pm 0.1^{c}$ | $1199 \pm 1^{d}$ | $120.2 \pm 37.7^{c}$ | $299 \pm 269^{*}$ | $112 \pm 73^{*}$ |

Different letters in the same column indicate significant difference (Holm-Sidak method at $P < 0.05$); DM: dry mass; NVG: no vegetative growth; $^{*}$no vegetative growth for *S. viminalis* at these levels of Cu treatment; values therefore represent that of *I. sachalinensis*. Values are mean ± standard deviation.

analysis of variance was carried out to evaluate the effects of treatments on the measured properties. In cases of significant difference ($P < 0.05$) all pair-wise comparison (Holm-Sidak multiple procedures) was done to identify the source that differs.

## 3. Results

*3.1. Soil Copper Content.* The $Cu_{\text{-TOT}}$ of the control agricultural soil (0 mg Cu $kg^{-1}$ soil) was 22 ± 6 mg $kg^{-1}$ soil and 16 ± 2 mg $kg^{-1}$ soil under *S. viminalis* and *I. sachalinensis*, respectively (Table 2). However, due to high variability, $Cu_{\text{-TOT}}$ was not significantly different under both plant species (Table 2). Upon treatment, $Cu_{\text{-TOT}}$ increased with increasing concentration of Cu applied (Table 2). Similarly, $Cu_{\text{-TOT}}$ showed high variability under both *S. viminalis* and *I. sachalinensis* at all levels of Cu treatment. Consequently, values were not significantly different under both plant species at each level of soil Cu treatment (Table 2). However, average $Cu_{\text{-TOT}}$ across plants showed significantly increased $Cu_{\text{-TOT}}$ with increasing concentration of Cu applied (Table 2). The $Cu_{\text{-PAV}}$ followed a similar trend; $Cu_{\text{-PAV}}$ generally increased with increasing $Cu_{\text{-TOT}}$ under both *S. viminalis* and *I. sachalinensis* (Table 2). Likewise, average $Cu_{\text{-PAV}}$ across plants revealed significantly increased $Cu_{\text{-PAV}}$ with increasing $Cu_{\text{-TOT}}$ (Table 2).

For the sewage amended soils, $Cu_{\text{-TOT}}$ was not significantly different under both plant species (Table 3). Values averaged 584 ± 44 mg $kg^{-1}$ soil and 569 ± 52 mg $kg^{-1}$ soil under *S. viminalis* and *I. sachalinensis,* respectively (Table 3), which were more than 25-fold compared to the control agricultural soils. The $Cu_{\text{-PAV}}$ of the sewage amended soils averaged 9.7 ± 0.8 mg $kg^{-1}$ soil and 9.1 ± 0.9 mg $kg^{-1}$ soil under *S. viminalis* and *I. sachalinensis*, respectively (Table 3). However, values were not significantly different between plant species (Table 3). After treatment of the sewage amended soils with EDTA, $Cu_{\text{-PAV}}$ increased to 63.2 ± 12.6 mg $kg^{-1}$ soil and 112.9 ± 27.1 mg $kg^{-1}$ soil under *S. viminalis* and *I. sachalinensis*, respectively (Table 3). This represents an increase of more than 6-fold in $Cu_{\text{-PAV}}$ compared to sewage amended soils without EDTA treatment (Table 3).

*3.2. Copper Accumulated in Stem and Leaf.* Increase in soil $Cu_{\text{-PAV}}$ with increasing $Cu_{\text{-TOT}}$ led to an increase in Cu content in the stem ($Cu_{\text{-STEM}}$) and leaf ($Cu_{\text{-LEAF}}$) of both

Table 3: Soil pH, total soil Cu ($\text{Cu}_{\text{-TOT}}$), available soil Cu ($\text{Cu}_{\text{-PAV}}$), and Cu accumulated in harvested stem ($\text{Cu}_{\text{-STEM}}$) and leaf ($\text{Cu}_{\text{-LEAF}}$) of *S. viminalis* and *I. sachalinensis* grown on sewage amended soils (SAMS) and SAMS treated with ethylenediaminetetraacetic acid (SAMS + EDTA).

| | pH [$\text{CaCl}_2$] | $\text{Cu}_{\text{-TOT}}$ [mg kg$^{-1}$ soil] | $\text{Cu}_{\text{-PAV}}$ [mg kg$^{-1}$ soil] | $\text{Cu}_{\text{-STEM}}$ [mg kg$^{-1}$ DM] | $\text{Cu}_{\text{-LEAF}}$ [mg kg$^{-1}$ DM] |
|---|---|---|---|---|---|
| Sewage amended soils | | | | | |
| *S. viminalis* | 6.9 ± 0.1$^a$ | 584 ± 44$^a$ | 9.7 ± 0.8$^a$ | 23 ± 4$^a$ | 30 ± 11$^a$ |
| *I. sachalinensis* | 6.8 ± 0.1$^a$ | 569 ± 52$^a$ | 9.1 ± 0.9$^a$ | 29 ± 8$^a$ | 17 ± 6$^a$ |
| SAMS + EDTA | | | | | |
| *S. viminalis* | 6.7 ± 0.3$^a$ | 651 ± 48$^a$ | 63.2 ± 12.6$^a$ | 55 ± 35$^a$ | 181 ± 121$^a$ |
| *I. sachalinensis* | 7.1 ± 0.4$^b$ | 630 ± 75$^a$ | 112.9 ± 27.1$^b$ | 195 ± 135$^b$ | 617 ± 474$^b$ |
| Soil treatment | | | | | |
| Sewage amended soils | 6.8 ± 0.1$^a$ | 576 ± 45$^a$ | 9.4 ± 0.8$^a$ | 26 ± 7$^a$ | 23 ± 11$^a$ |
| SAMS + EDTA | 6.9 ± 0.4$^a$ | 641 ± 59$^b$ | 88.0 ± 33.0$^b$ | 125 ± 118$^b$ | 399 ± 396$^b$ |

Different letters in the same column indicate significant difference (Holm-Sidak method at $P < 0.05$). DM: dry mass. Values are mean ± standard deviation.

*S. viminalis* and *I. sachalinensis* (Table 2). The $\text{Cu}_{\text{-STEM}}$ and $\text{Cu}_{\text{-LEAF}}$ of *S. viminalis* grown on agricultural soils treated with 250 mg Cu kg$^{-1}$ soil were comparable to those of the control (Table 2). At Cu treatment beyond 250 mg Cu kg$^{-1}$ soil, there was no vegetative growth in *S. viminalis* (Table 2). Interestingly, *I. sachalinensis* continue to grow and accumulate Cu in the stem and leaf at higher soil Cu content (Table 2).

For plants grown on sewage amended soils, $\text{Cu}_{\text{-STEM}}$ of *S. viminalis* and *I. sachalinensis* averaged 23 ± 4 [mg kg$^{-1}$ DM] and 29 ± 8 [mg kg$^{-1}$ DM], respectively, whereas $\text{Cu}_{\text{-LEAF}}$ was 17 ± 6 [mg kg$^{-1}$ DM] for *S. viminalis* and 30 ± 11 [mg kg$^{-1}$ DM] for *I. sachalinensis*; however, values were not significantly different between plant species (Table 3). The enhanced $\text{Cu}_{\text{-PAV}}$ after treatment of sewage amended soils with EDTA led to significant increase in $\text{Cu}_{\text{-STEM}}$ and $\text{Cu}_{\text{-LEAF}}$ of *I. sachalinensis* compared to *S. viminalis* (Table 3).

Accumulated $\text{Cu}_{\text{-STEM}}$ and $\text{Cu}_{\text{-LEAF}}$ of *S. viminalis* and *I. sachalinensis* grown on sewage amended soils were about 250% higher in comparison to the control agricultural soils. However, upon treatment with EDTA, $\text{Cu}_{\text{-STEM}}$ and $\text{Cu}_{\text{-LEAF}}$ of *S. viminalis* and *I. sachalinensis* increased to around 1000% and 3100%, respectively, compared to the control agricultural soils.

## 4. Discussions

At lower soil Cu content, both *S. viminalis* and *I. sachalinensis* accumulated comparable amounts of Cu in the stem and leaf. Copper accumulated in the leaf of *S. viminalis* grown on the control agricultural soil and that treated with 250 mg Cu kg$^{-1}$ soil was within the tolerable range of 3 and 15 mg kg$^{-1}$ as reported by Blume [23] whereas that of the stem was well within the critical range of 15 and 20 mg kg$^{-1}$ [23]. However, it appears that such threshold values for *I. sachalinensis* are currently lacking in the literature. For the agricultural soils treated with Cu, generally more Cu accumulated in the stem than in the leaf, especially for *I. sachalinensis*. Compared to other heavy metals, Cu is known to be less mobile in plant tissues [16, 19, 24]. According to Nissen and Lepp [24], Punshon and Dickinson [19], and Kuzovkina et al. [16], most of the Cu is usually immobilized in the roots of *S. viminalis*. However, in this study Cu accumulation in the belowground tissues was not investigated. As noted by Baker and Brooks [25], plants have evolved adaptive strategies in response to heavy metal stress: ability to detoxify metal ions at different location and transport them to the shoot where they can be stored in vacuoles of leaf cells [25].

The observed ability of *S. viminalis* and *I. sachalinensis* to continue growth in the presence of Cu and to accumulate Cu in their tissues demonstrates their tolerance to moderate-to-high levels of Cu contamination, especially for *I. sachalinensis*. Copper accumulated in the stem and leaf varied between the two plant species with increasing soil Cu concentration. This may be due to the difference in sensitivity between species, ranging from the stimulation of growth to severe inhibition [16]. Thus, the lack of vegetative growth in *S. viminalis* at Cu treatments exceeding 250 mg Cu kg$^{-1}$ soil is presumably a result of soil Cu reaching levels phytotoxic to *S. viminalis*. This seems to suggest that *S. viminalis* is relatively less resilient to soil Cu content. On the other hand, the continuous vegetative growth and Cu accumulation in the stem and leaf of *I. sachalinensis* at higher concentrations suggest that *I. sachalinensis* is more resilient and has the potential to tolerate and phytoextract Cu from soils with elevated Cu content.

Copper accumulated in the stem and leaf showed a similar trend for both *S. viminalis* and *I. sachalinensis* grown on sewage amended soils. However, treatment with EDTA greatly enhanced the availability of soil Cu and consequently led to increased Cu accumulation in the leaf and stem of *S. viminalis* and *I. sachalinensis* with values exceeding the tolerable range reported by Pulford et al. [21]. This may be attributed to enhanced mobility of soil Cu by EDTA and subsequent uptake and translocation by the plants to the aboveground tissues. As noted by Yang et al. [26], Cu uptake by plant shoots is directly related to the soluble Cu in the soil solum [26]. However, EDTA is known to form biochemically stable complexes with heavy metals that are not readily degradable and may persist in the soil environment [11, 12]. Excessive application of EDTA may therefore lead to residual toxicity and eventually seep into groundwater bodies [10].

## 5. Conclusions

Treatment of agricultural soils with increasing levels of Cu concentration led to increased accumulated Cu in the stem and leaf of *S. viminalis* and *I. sachalinensis*. Addition of EDTA to sewage amended soils enhanced Cu available and increased Cu accumulated in the stem and leaf of the plant species. *Salix viminalis* and *I. sachalinensis* grown on long-term field sewage amended soils with large background total Cu content accumulated higher Cu in stem and leaf compared to those grown on the control agricultural soils with low background Cu content. The ability of *I. sachalinensis* to grow at higher soil Cu content and accumulate larger amounts of Cu in the stem and leaf suggests that *I. sachalinensis* is relatively more resilient and could tolerate elevated soil Cu content. *Igniscum sachalinensis* therefore seems to have greater potential for phytoremediation of Cu-contaminated soils compared to *S. viminalis*. However, to establish its true potential for phytoextraction of Cu, further field test is needed since plant behaviour and performance in field metal-contaminated soil may differ from that under greenhouse conditions.

## Conflict of Interests

The authors declare that they have no conflict of interests regarding the publication of this paper.

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