The Potential of Zambian Copper-Cobalt Hyper-accumulator Plants for Phytoremediation of polluted (Mining/Smelter) Soils

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Key themes: Community and environmental sustainability
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Research aims:
This project focused on Cu-Co hyperaccumulator plants occurring in the Copper-Cobalt Belt in Zambia, to unlock their potential for phytoextraction to offer a low-cost approach to rehabilitate metal-contaminated soils.
The Action Research aimed to elucidate metal speciation and elemental distribution in selected Cu-Co hyperaccumulator plants with high potential for phytoextraction.

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Action Research Report

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Summary of Action Research Activity

The potential of Zambian Copper-Cobalt hyper-accumulator plants for phytoremediation of polluted (mining/smelter) soils

The Copper-Cobalt Belt of the Democratic Republic of Congo and Zambia is one of the most important metallogenic regions and, without doubt, the richest metallophyte (plants endemic to metal-enriched soils) location in the world. The existence of more than 30 Cu-Co hyperaccumulator plants is a special feature of this region and these plants accumulate extraordinarily high concentrations of Cu and Co metal in their living tissues. Such plants have the potential to be used for phytotechnologies; i.e. by growing and harvesting these plants, Cu-Co can be removed from (polluted) soils. This process can serve to remediate contaminated soils, for example around smelters (phytoextraction), or create a 'metal enriched crop' (phytomining). This project aimed to elucidate metal speciation and elemental distribution in selected Cu-Co hyperaccumulator plants with high potential for phytoextraction.

The fieldwork campaign was successfully completed in Zambia in October–November 2014. During the fieldwork the team visited a range of active and abandoned mines and tailings storage facilities in the Copperbelt Region of Zambia. The team also visited two First Quantum Minerals (FQML) mine sites: Bwana Mkubwa and Kansanshi, and the Copperbelt University in Kitwe. During the fieldwork a range of metallophytes and Cu-Co hyperaccumulator plants were discovered. The team collected approximately 200 plant specimens, 25 soil samples and 10 mineral samples for chemical analysis. Cryogenically preserved samples from 3 different Cu-Co hyperaccumulator species were also collected for advanced analysis in South Africa and Australia. The micro-PIXE analysis on the collected plant samples took place in May 2015 in South Africa, and synchrotron analysis in Australia is scheduled for July 2015.

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THE POTENTIAL OF ZAMBIAN COPPER-COBALT HYPERACCUMULATOR PLANTS FOR PHYTOREMEDIATION OF POLLUTED (MINING/SMELTER) SOILS

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Acknowledgements

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Glossary

AMD (Acid and Metalliferous Drainage) – contaminated liquid produced from the interaction of rock minerals with water. AMD is typically formed through the decomposition of sulphide minerals upon exposure to atmospheric conditions and contains dissolved metals and other minor elements that can be environmentally harmful.

Metallophyte – plant species that has evolved specific tolerance to prevailing high trace elements concentrations in the soil in which it grows**.**

Phytoextraction – technology of using hyperaccumulator plants to extract unwanted trace elements from the soil or minerals waste with the aim of remediating pollution.

Phytomining – using hyperaccumulator plants to extract target trace elements from the soil or minerals waste with the aim of recovering such elements for profit.

Reclamation – reclaiming land for a productive purpose (e.g. food crops) including monoculture plantations.

Recolonisation – re-establishment of plants that are self-sown or by vegetative regrowth.

Rehabilitation – the process of improving disturbed land, including the development of new plant communities that may include some of the original plant species and may, or may not, have economic outcomes.

Restoration – recreating an ecological community that existed prior to clearing, mining or other disturbance.

Tailings – materials left over after separating the valuable fraction from the uneconomic fraction of an ore following mineral processing.

Waste Rock – uneconomic rock extracted during mining that has not been processed.

Introduction

The Copper-Cobalt Belt of the Democratic Republic of the Congo and Zambia is one of the most important metallogenic regions and, without doubt, the richest metallophyte (plants endemic to metal-enriched soils) location in the world. To date, over 600 metallophytes have been described from this region, including many species that occur nowhere else. These metallophytes occur on isolated copper-mineralized hills spread throughout the Copper-Cobalt Belt. The existence of more than 30 Cu-Co (copper-cobalt) hyperaccumulator plants is a special feature of this region, and these plants accumulate extraordinarily high concentrations (up to 2%) of Cu and Co metal in their living tissues. Such hyperaccumulators plants have the potential to be used for phytotechnologies; i.e. by growing and harvesting these plants Cu-Co can be removed from (polluted) soils. This process can serve to remediate contaminated soils, for example around smelters (phytoextraction), or create a 'metal enriched crop' (phytomining). The concomitant occurrence of Cu-Co hyperaccumulator plants and large areas with metal-enriched and metal contaminated soils makes a compelling case for developing these phytotechnologies in Zambia. In addition, Cu-Co metallophytes originating from naturally mineralized areas may colonize industrial minerals wastes and mined-out areas and as such establish vegetative cover and contribute to rehabilitation.

Research on Cu-Co metallophytes from the Copper-Cobalt Belt has almost exclusively focused on the DR Congo, mainly by the academics of the Université Libre de Bruxelles and Université de Liège Gembloux in Belgium (http://copperflora.org/). The focus of the research by these teams has been on plant taxonomy, vegetation and conservation. Several publications have now also emphasized that many of the unique metallophytes of the Copper-Cobalt Belt are under significant threat as a result of widespread mining activities and habitat-destruction. Action towards conserving the metallophyte resource of the Copper-Cobalt Belt is therefore imperative and represents a great asset to the mining industry in DR Congo and Zambia. Although the phenomenon of abnormal accumulation of Cu-Co in certain plants from the Copper-Cobalt Belt has been known since the 1950s, no study to date has elucidated metal speciation and distribution in these Cu-Co hyperaccumulator plants. In fact, a recent study (in the DR Congo) has shown that most earlier plant analyses were contaminated and that, whilst still hyperaccumulating Cu-Co, the true extent of this phenomenon is unclear. This may be explained by the lack of availability or access to suitable methods to analyse metals *in situ* in plant tissues (synchrotron and micro-probe techniques), which are now available to be used in this project.

Even though successful phytotechnologies have been demonstrated for nickel elsewhere in the world, this project instigates the first Cu-Co hyperaccumulator phytotechnologies to have ever been developed. The Action Research Project initiated screening and analysis of metal distribution of Cu-Co hyperaccumulator plants located in the natural habitat in Zambia.

Methods

Field assessment

Our fieldwork was conducted in late October and early November 2014 in areas suggested by research team members, François Malaisse, Royd Vinya and Kenneth Maseka, that might contain species of the copper-cobalt flora. We assessed a large number of sites (see Figure 1) covering a large portion of the Copperbelt region in northern Zambia. We assessed 'Zambian type anomalies' including Bwana Mkubwa and Roan Antelope, potential areas near Kitwe and Ndola and 'Katangan type outcrops' of Kansanshi near the town of Solewzi. Representative flora were collected and identified at these sites.

Collection and elemental analysis of plant tissues

Plant tissue samples of *Conyza cordata* (Asteraceae), *Persicaria puncata* and *Persicaria capitata* (Polygonaceae) were collected along a stream polluted by copper/cobalt slag near Kitwe. From each species tissue samples were collected from the following parts: roots, stem and leaves. The tissue samples were cut out with a surgical knife directly from the living plant. The samples were (in the field) immediately flash-frozen using a cold mirror technique in which the samples were pressed against a large block of copper metal cooled by liquid nitrogen (-196°C) and a second block of attached to a Teflon holder. This ensured extremely fast freezing of the plant tissue samples to prevent cellular damage by ice crystal formation. Parallel specimens were collected and fixed in 3% gluteraldehyde for anatomical studies. The samples were then transported on a cryogenic container directly to Australia and South Africa for analysis.

Collection and analysis of soil samples

Soil samples were also collected between the roots of *Conyza cordata* (Asteraceae), *Persicaria puncata* and *Persicaria capitata* (Polygonaceae). The soil samples were air-dried at room temperature to constant weight, sieved to <2 mm, a fraction of each soil sample (10 g) was re-dried at 105°C for 48h to allow for moisture corrections. The soil samples (300 mg) were digested using freshly prepared Aqua Regia (4 ml 70% nitric acid and 3 ml 37% hydrochloric acid per sample) in a digestion block for 2 hours, diluted to 10 ml before analysis. Soil moisture pH and electrical conductivity (EC) was obtained in a 1:2.5 soil: water mixture. Diethylene triamine pentaacetic acid (DTPA) extraction was used to determine the various pools of metals in the soil. All samples were analysed with ICP-AES (Varian Vista Pro) for Ni, Co, Cu, Zn, Mn, Pb, Cd, Fe, Mg, Ca, Na, K, S and P.

Collection of plant tissues and preparation for micro-PIXE analysis

At iThemba LABS, samples for anatomical studies were post-fixed in 2% osmium tetraoxide (OsO4), dehydrated in a graded, ethanol series and embedded in Spurr's (1969) resin. Sections 0.5–2μm thick were cut with a diamond knife, stained with Azur II and methylene blue, examined and photographed with a compound microscope.

In iThemba LABS cryo-laboratory, the frozen specimens were removed from the LN_2 storage container and were lyophilized in a Leica EM CFD Cryosorption Freeze Dryer (Leica Microsystems AG, Austria The program used started at -80°C and ended at room temperature about 210h later. The long freeze-drying cycle is used in order to avoid shrinkage. Sectioned specimens were mounted on aluminium target frames between two layers of Formvar® film (0.5% Formvar®). To make the surface layer conductive and prevent charge build-up during analysis, the Formvar® films had been previously carbon coated. All specimens were photographed under stereomicroscope using a digital camera before and after analysis.

Micro-PIXE elemental mapping (undertaken)

Microanalyses were performed using the nuclear microprobe at the Materials Research Department, iThemba LABS, South Africa. The facility and methodology of measurements of biological materials have been described by Prozesky *et al.*, 1995; Przybyłowicz *et al.*, 1999; Przybyłowicz *et al.*, 2005. A proton beam of 3 MeV energy, provided by the 6 MV single-ended Van de Graaff accelerator, was focused to a 3 x 3 μm^2 spot and raster scanned over the areas of interest, using square or rectangular scan patterns with a variable number of pixels (up to 128 x 128). Proton current was restricted to 100 - 150 pA to minimize specimen beam damage. Particle-induced X-ray emission (PIXE) and proton backscattering spectrometry (BS) were used simultaneously. PIXE spectra were registered with a Si(Li) detector manufactured by PGT (30 mm^2 active area and 8.5 μm Be window) with an additional 125 μm Be layer as an external absorber. The effective energy resolution of the PIXE system (for the Mn Kα line) was 160 eV, measured for individual spectra. The detector was positioned at a takeoff angle of 135^o and a working distance of 24 mm. The X-ray energy range was set between 1 and 40 keV. BS spectra were recorded with an annular Si surface barrier detector (100 μm thick) positioned at an average angle of 176°. Data were acquired in the event-by-event mode.

Quantitative results were obtained by standardless method using GeoPIXE II software package (Ryan *et al.*, 1990a; Ryan *et al.*, 1990b; Ryan, 2000). The error estimates are extracted from the error matrix generated in the fit, and the minimum detection limits (MDL) are calculated using the Currie equation (Currie 1968). The detailed calibration of detector efficiency, the thicknesses of selectable x-ray attenuating filters and studies on the accuracy and precision have been reported elsewhere (van Achterbergh *et al*. 1995). The calibration of the analytical system was tested by measurements of standards - pure elements and synthetic glasses with known quantities of selected minor elements (internal standards), the x-ray peaks of which

cover practically the whole measurable energy range. Quantitative elemental mapping was performed using *Dynamic Analysis* method (Ryan & Jamieson, 1993; Ryan *et al.*, 1995; Ryan, 2000). This method generates elemental images, which are (i) overlap-resolved, (ii) with subtracted background and (iii) quantitative, i.e. accumulated in mg kg^{-1} dry weight units. Maps were complemented by data extracted from arbitrarily selected micro-areas within scanned plant tissue. PIXE and BS spectra were employed to obtain average concentrations from these micro-areas using a full nonlinear deconvolution procedure to fit PIXE spectra (Ryan *et al.*, 1990a; Ryan *et al.*, 1990b), with matrix corrections based on thickness and matrix composition obtained from the corresponding BS spectra, fitted with a RUMP simulation package (Doolittle 1986) with non-Rutherford cross-sections for C, O, N. Elemental concentrations from these areas are also reported in mg kg^{-1} dry weight.

Synchrotron micro-XRF and XANES mapping (not yet undertaken)

The frozen samples are sectioned using a cryo-microtome to 40–60 µm. The frozen-hydrated state of the samples provides optimum conditions for µXRF and XANES imaging. The width of the N_2 cold cone of the cryo-stream is approximately 15 mm, depending on the distance to the sample when set to –100°C.

Two consecutive measurements are conducted for the samples: the first scan represents a µXRF map, the second scan fluorescence-XANES imaging. The tissues portions/sections are to be film-sandwiched between two sheets of Kapton 4 µm thin film held in suspension between a Perspex U-frame on the motorised sample stage of the XFM in front of the Maia detector under the N_2 cryo-stream. The bulk elemental concentrations in the samples intended for the XFM experiment are in the range of 100–200 µg g^{-1} (Cu) and 300–500 µg g^{-1} (Co). Two replicate samples from leaf portions of the three species will be imaged using the KB/Maia setup with a 25 ms/pixel dwell time.

Fluorescence XANES imaging will be performed to obtain laterally-resolved speciation of Cu and Co in the leaves. The XANES imaging consists of forming ‘stacks’ of µXRF maps (corresponding to a series of 50 energies across the Cu and Co K-edges, each separately) while progressively increasing the energy. We will compare the XANES spectra against model compound spectra and bulk XAS which have previously been recorded at the Australian Synchrotron.

Main findings

The fieldwork enabled the team to collect samples from a number of populations of hyperaccumulator plants in the Zambian Copperbelt. However, the fieldwork also made it evident that the high rates of human-induced disturbances due to industrial mining has led to major losses in the metallophytes and hyperaccumulator plant diversity. The subsequent artisanal mining activities have without doubt resulted in further loss of the plant populations that once thrived under the local ecological settings. The time assigned to this Action Research was insufficient to cover the wider Zambian metalliferous region and to gain a better understanding of the occurrences of hyperaccumulators in Zambia. For long-term sustainability of this project, the engagement of two research students at Ph.D. level will be an appropriate strategy. The many subjects students could focus on include: (i) the ecophysiology of copper-cobalt hyperaccumulator plants; (ii) *ex-situ* conservation of hyperaccumulator plants and metallophytes; and (iii) nursery-based experimental research.

In summary, according to our field survey, it appears that:

1. The original flora on natural copper-cobalt mineralised sites has been destroyed in all places that have been subjected to mining. This is the case for: (i) “Katangan type outcrops” (namely Kansanshi) and (ii) the “Zambian type anomalies” (a large number of sites including Bwana Mkubwa and Roan Antelope, Kitwe, etc.).

2. Untouched “Zambian type anomalies” still exist in some places (judging from photographs available locally). It is, therefore, urgently necessary, both to identify these sites and to collect plant material from these sites.

3. Minerals waste dumps are of interest to sample local “weeds” that have been able to colonize these man-made polluted sites.

The project has established a partnership between the Copperbelt University, iThemba LABS and the University of Queensland on mine/contaminated site rehabilitation so that the Zambia partners have a network of international research partners to advance phytotechnologies locally. We have also shared methods of field screening and trained some of our partners in cutting-edge nuclear micro-probe (micro-PIXE) and synchrotron analysis to elucidate Cu-Co metal speciation and distribution in the plant tissues.

The team members are current developing strategies for acquiring appropriate funding to support more detailed follow-up studies. This includes negotiations with mining companies active in the Copperbelt, and grant applications from government agencies.

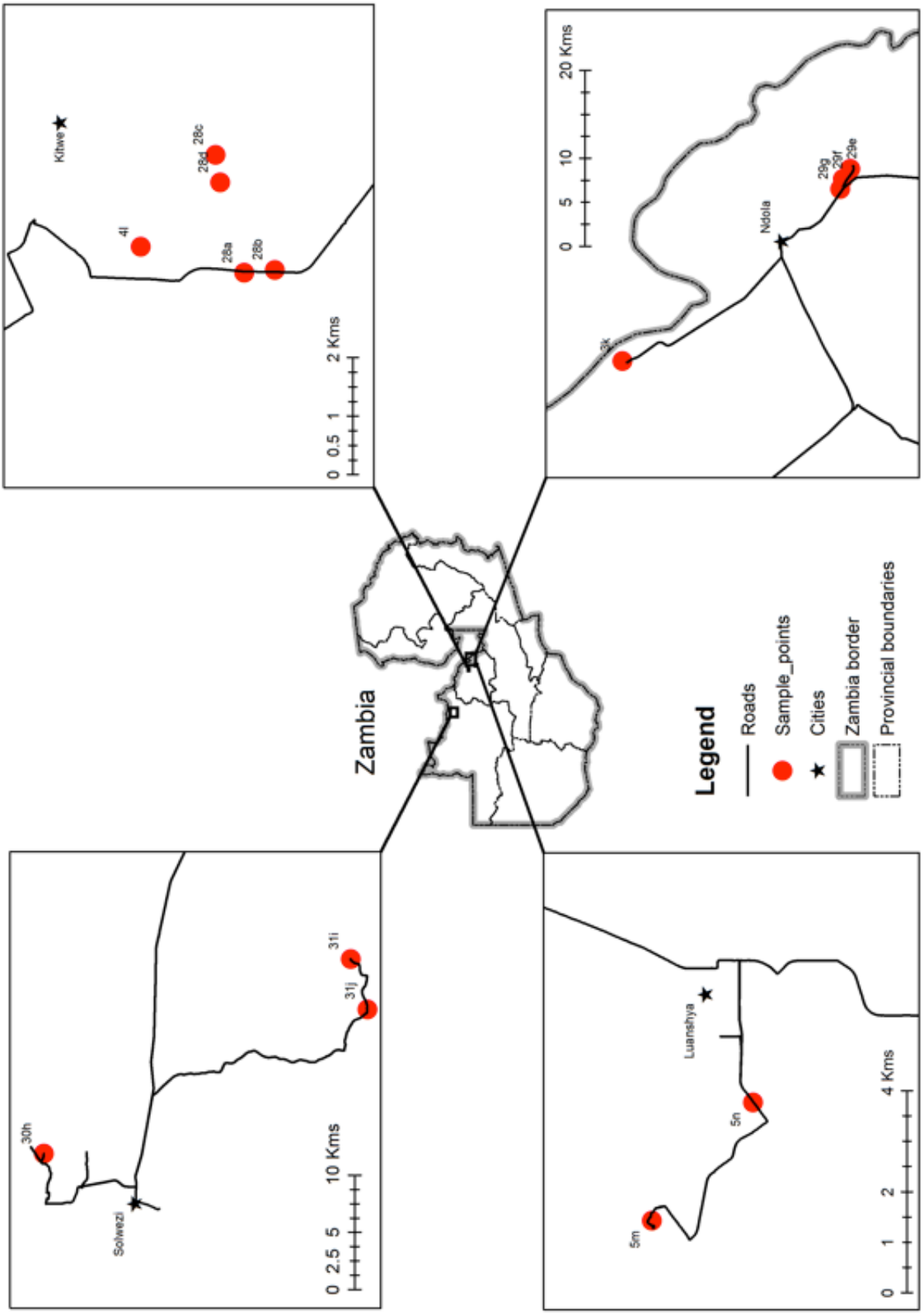

Figure 1. Map showing the localities and sites visited during the fieldwork in Zambia.

Figure 2. The team in the field at the Kansanshi Mine site in Northern Zambia.

Figure 3. Together with collaborators from the Copperbelt University studying herbarium specimens at the Forest Department in Ndola.

Figure 4. Visit to the Copperbelt University chemistry classes.

Figure 5. The team at the micro-PIXE beamline of iThemba LABS conducting the experiments.

Figure 6. *Conyza cordata*, one of the Cu hyperaccumulating plants collected during the fieldwork.

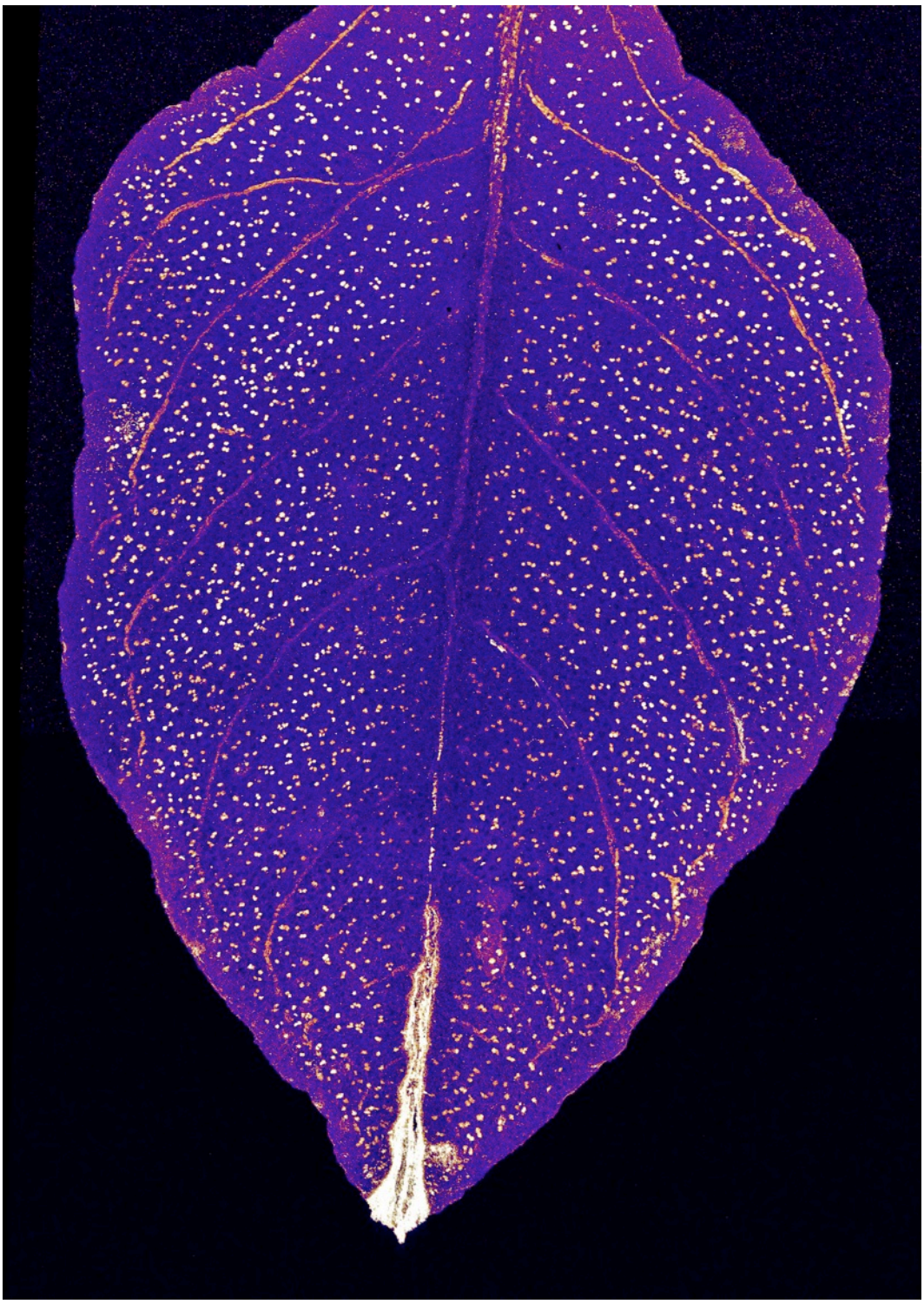

Figure 7. Preliminary synchrotron XFM image of a *Conyza cordata* leaf.

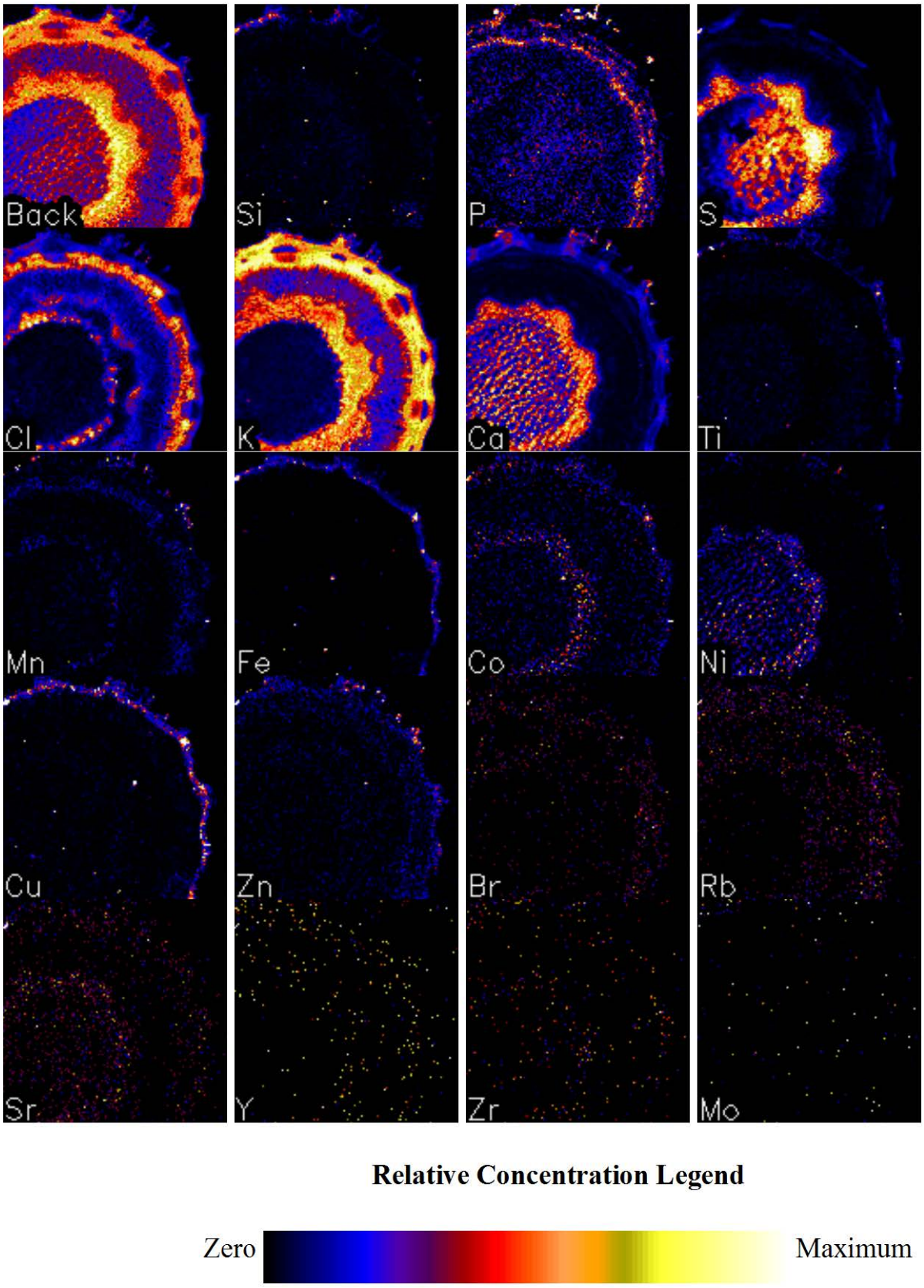

Figure 8. Preliminary micro-PIXE data showing quantitave elemental maps of *Conyza cordata* stem of is presented below. For each plant species 3 specimens of root, stem and leaf were analyzed.

Table 1. Preliminary identifications of collected herbarium specimens from the sites.

Refer.	Site	Family	Name
28 A	Ushi dam, West Aka 16ft bridge	Campanulaceae	*Lobelia erinus* L.
28 A	Ushi dam, West Aka 16ft bridge	Polygonaceae	*Persicaria capitata* (Buch.-Ham. ex D.Don) H.Gross
28 A	Ushi dam, West Aka 16ft bridge	Asteraceae	*Anisoppapus chinensis* (L.) Hook & Arn. subsp. *chinensis*
28 A	Ushi dam, West Aka 16ft bridge	Convolvulaceae	*Ipomoea* sp.
28B	Ushi dam, West Aka, stream	Pteridaceae	*Pteris vittata* L.
28B	Ushi dam, West Aka, stream	Asteraceae	*Conyza pyrhopappa* A.Rich.
28B	Ushi dam, West Aka, stream	Portulacaceae	*Portulacca* sp.
28B	Ushi dam, West Aka, stream	Cyperaceae	*Cyperus dives*
28B	Ushi dam, West Aka, stream	Amaranthaceae	*Celosia trigyna* L.
28C	Uchi East, tailing	Polygonaceae	*Persicaria* sp.
28C	Uchi East, tailing	Poaceae	*Setaria* sp.
28C	Uchi East, tailing	Poaceae	*Loudetia* sp.1
28C	Uchi East, tailing	Poaceae	*Loudetia* sp.2
28C	Uchi East, tailing	Poaceae	*Cymbopogon densiflorus*
28C	Uchi East, tailing	Poaceae	*Hyparrhenia* sp.
28C	Uchi East, tailing	Cyperaceae	*Bulbostylis* sp.
28C	Uchi East, tailing	Amaranthaceae	*Celosia* sp.
28C	Uchi East, tailing	Asteraceae	*Pseudognaphalium luteo-album*
28D	Uchi, stream	Myrophyllaceae	*Myriophyllum* sp.
29E	Bwana Mkubwa, Rompod mine	Cyperaceae	*Bulbostylis pseudoperennis*
29E	Bwana Mkubwa, Rompod mine	Convolvulaceae	*Ipomoea cairica* (L.) Sweet var. *cairica*
29E	Bwana Mkubwa, Rompod mine	Convolvulaceae	*Ipomoea* sp.1
29E	Bwana Mkubwa, Rompod mine	Convolvulaceae	*Ipomoea* sp.1
29E	Bwana Mkubwa, Rompod mine	Asteraceae	*Tridax procumbens*
29E	Bwana Mkubwa, Rompod mine	Asteraceae	*Pseudognaphalium luteo-album* (L.) Hilliard & Burtt
29E	Bwana Mkubwa, Rompod mine	Ochnaceae	*Ochna* sp.
29E	Bwana Mkubwa, Rompod mine	Fabaceae	*Tephrosia* sp.
29E	Bwana Mkubwa, Rompod mine	Borraginaceae	*Trichodesma* sp.
29E	Bwana Mkubwa, Rompod mine	Poaceae	*Arthraxon quartinianus*
29F	Old concentrator, facing station	*n.a.*	*Pityrogramma calomelanos* (SW.) Link var. *aureoflava* (Hook.) Weath. ex Bailey
29F	Old concentrator, facing station	Phormidiaceae	*Pyrphyrosiphon notarsii* Kütz. ex Gomont
29F	Old concentrator, facing station	Apiceae	*Diplolophium* sp.
29G	Mine pit summit	Asteraceae	*Vernonia* sp.
29G	Mine pit summit	Fabaceae	*Abrus* sp.

29G	Mine pit summit	Anacardiaceae	*Rhus* sp.
29G	Mine pit summit	Asteraceae	*Vernonia* sp. 2
29G	Mine pit summit	Rubiaceae	cf. *Spermacoce*
30H	South East Dome	Lamiaceae	*Becium centraliafricanum* (R.E.Fries) Sebald
30H	South East Dome	Orobanchaceae	*Buchnera henriquesii* Engl.
30H	South East Dome	Rubiaceae	*Fadogia cienkowski* Schweinf.
30H	South East Dome	Tiliaceae	*Triumfetta digitata* (Oliv.) Sprague
30H	South East Dome	Annonaceae	*Annona stenophylla* Engl. & Diels subsp. *nana* (Exell) N.Robson
30H	South East Dome	Fabaceae	*Erisosema engleriana* Harms
30H	South East Dome	Commelinaceae	*Cyanotis* sp.
31I	miombo to pit	Euphorbiaceae	*Acalypha*
31I	miombo to pit	Lamiaceae	*Becium*
31I	miombo to pit	Cucurbitaceae	*Trochomeria macrocarpa* (Sond.) Hook.
31I	miombo to pit	Aristolochiaceae	*Aristolochia heppii* Merxm.
31J	river(stream) Mwindishi	Aponogetonaceae	*Aponogeton desertorum*
31I	miombo to pit	Malavaceae	*Hibiscus rhodanthus*
3L	Mwekera	Passifloraceae	*Adenia lobata* (Jacq.) subsp. *rumicifolia* (Engl. & Harms) Lye
3L	Mwekera	Asparagaceae	*Asparagus africanus* Lam. var. africanus
3L	Mwekera	Vitaceae	*Cyphostemma*
3L	Mwekera	Rubiaceae	*Fadogia cienkowski* Schweinf.
3L	Mwekera	Mimosaceae	*Albizia adianthifolia* (Schumach.) W.Wight
3L	Mwekera	Smilacaceae	*Smilax anceps* Willd.
3L	Mwekera	Campanulaceae	*Wahlenbergia* sp.
3L	Mwekera	Passifloraceae	*Adenia gummifera* (Harv.) Harms var. *gummifera*
3L	Mwekera	Commelinaceae	*Commelina* sp.
3L	Mwekera	Convolvulaceae	*Ipomoea* sp.
3L	Mwekera	Cucurbitaceae	*Trochomeria macrocarpa* (Sond.) Hok.
3L	Mwekera	Loganiaceae	*Strychnos spinosa* Lam.
3L	Mwekera	Acanthaceae	*Thunbergia* sp.
3L	Mwekera	Commelinaceae	*Cyanotis* sp.
3L	Mwekera	Fabaceae	*Tephrosia* sp.

Table 2. Preliminary plant material analytical results from XRF measurements.

Refer.	Family	Name	Se	As	Hg	Zn	Cu	Ni	Co
28 A	Orobanchaceae	*Alectra sessiliflora* (Vahl) Kuntze	0	0	0	918	349	0	0
28 A	Asteraceae	*Anisoppapus chinensis* (L.) Hook & Arn. subsp. *chinensis*	0	0	0	485	655	0	128
28 A	Convolvulaceae	*Ipomoea* sp.	0	0	0	110	370	0	0
28 A	Campanulaceae	*Lobelia erinus* L.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
28 A	Polygonaceae	*Persicaria capitata* (Buch.-Ham. ex D.Don) H.Gross	0	0	0	254	1306	0	0
28B	Amaranthaceae	*Celosia trigyna* L.	0	0	0	148	723	0	1060
28B	Asteraceae	*Conyza pyrhopappa* A.Rich.	0	0	0	238	1284	0	592
28B	Cyperaceae	*Cyperus dives*	128	0	0	169	361	0	0
28B	Portulacaceae	*Portulacca* sp.	0	0	0	15	59	0	0
28B	Pteridaceae	*Pteris vittata* L.	0	259	0	69	374	0	0
28C	Cyperaceae	*Bulbostylis* sp.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
28C	Amaranthaceae	*Celosia* sp.	6	0	0	59	541	0	0
28C	Poaceae	*Cymbopogon densiflorus*	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
28C	Poaceae	*Hyparrhenia* sp.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
28C	Poaceae	*Loudetia* sp.1	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.	n.a.
28C	Poaceae	*Loudetia* sp.2	4	0	0	0	146	0	0
28C	Polygonaceae	*Persicaria* sp.	5	0	0	85	697	0	208
28C	Asteraceae	*Pseudognaphalium luteo-album*	0	0	0	122	1042	0	0
28C	Poaceae	*Setaria* sp.	n.a.	n.a.	n.a.	n.a.	n.a.	0	n.a.
28D	Myrophyllaceae	*Myriophyllum* sp.	n.a.	n.a.	n.a.	n.a.	n.a.	0	n.a.
29E	Poaceae	*Arthraxon quartinianus*	n.a.	n.a.	n.a.	n.a.	n.a.	0	n.a.
29E	Cyperaceae	*Bulbostylis pseudoperennis*	n.a.	n.a.	n.a.	n.a.	n.a.	0	n.a.
29E	Convolvulaceae	*Ipomoea cairica* (L.) Sweet var. *cairica*	0	0	0	99	368	0	0
29E	Convolvulaceae	*Ipomoea* sp.1	0	0	0	77	161	0	0
29E	Convolvulaceae	*Ipomoea* sp.1	0	0	0	39	1169	0	0
29E	Ochnaceae	*Ochna*	0	0	0	87	245	0	0
29E	Asteraceae	*Pseudognaphalium luteo-album* (L.) Hilliard & Burtt	0	0	0	265	738	0	0
29E	Fabaceae	*Tephrosia*	0	0	0	128	132	0	0
29E	Borraginaceae	*Trichodesma*	0	0	0	90	274	0	0
29E	Asteraceae	*Tridax procumbens*	0	0	0	458	386	0	0
29F	Apiceae	cf. *Diplolophium* sp.	0	0	0	206	3038	0	0

29F	n.a.	*Pityrogramma calomelanos* (SW.) Link var. *aureoflava* (Hook.) Weath. ex Bailey	0	46	0	111	283	0	0
29F	Phormidiaceae	*Pyrphyrosiphon notarsii* Kütz. ex Gomont	n.a.	n.a.	n.a.	n.a.	n.a.	0	n.a.
29G	Fabaceae	*Abrus*	0	0	0	111	325	0	0
29G	Rubiaceae	cf. *Spermacoce*	0	0	0	165	139	0	0
29G	Anacardiaceae	*Rhus*	0	0	0	38	231	0	0
29G	Asteraceae	*Vernonia*	11	0	0	296	332	0	0
29G	Asteraceae	*Vernonia* sp. 2	0	0	0	678	860	0	0
30H	Annonaceae	*Annona stenophylla* Engl. & Diels subsp. *nana* (Exell) N.Robson	0	0	0	49	186	0	0
30H	Lamiaceae	*Becium centraliafricanum* (R.E.Fries) Sebald	0	0	0	92	290	0	0
30H	Orobanchaceae	*Buchnera henriquesii* Engl.	0	0	0	88	184	0	0
30H	Commelinaceae	*Cyanotis* sp.	0	0	0	43	116	0	0
30H	Fabaceae	*Erisosema engleriana* Harms	0	0	0	124	559	0	0
30H	Rubiaceae	*Fadogia cienkowski* Schweinf.	0	0	0	51	315	0	0
30H	Tiliaceae	*Triumfetta digitata* (Oliv.) Sprague	0	0	0	95	315	0	0
31I	Euphorbiaceae	*Acalypha*	0	0	0	120	212	0	0
31I	Euphorbiaceae	*Acalypha*	0	0	0	29	156	0	0
31I	Euphorbiaceae	*Acalypha*	0	0	0	43	115	0	0
31I	Aristolochiaceae	*Aristolochia heppii* Merxm.	0	0	0	123	143	0	0
31I	Lamiaceae	*Becium*	0	0	0	264	194	0	0
31I	Fabaceae	*Crotalaria*	0	0	0	46	148	0	0
31I	Malavaceae	*Hibiscus rhodanthus*	0	0	0	227	201	0	0
31I	Fabaceae	*Indigofera*	0	0	0	78	137	0	0
31I	Cucurbitaceae	*Trochomeria macrocarpa* (Sond.) Hook.	0	0	0	45	170	0	0
31J	Aponogetonaceae	*Aponogeton desertorum*	n.a.	n.a.	n.a.	n.a.	n.a.	0	n.a.

Table 3. Preliminary soil analytical results from DTPA-extracts (indicative of plant-available metal concentrations).

Type	Location/species	Co	Cu	Fe	Mn	Ni	Zn
top soil	Tailings Luanshi	0.2	150	5.6	14	0.5	0.8
top soil	Tailings 4L	4.7	118	7	10	0.2	0.6
top soil	Roan Antilope waste dump	0.2	168	6.3	9	0.2	0.9
top soil	Tailings 28C	6.5	187	12	5	0.1	0.9
Rhizosphere	*Persicaria capitata*	4	378	30	5	0.2	6
Rhizosphere	*Conyza cordata*	8.3	256	8.3	3.8	0.1	2.1
Rhizosphere	*Persicaria puncata*	90	343	10	6.2	1	12
Rhizosphere	*Conyza cordata*	16	639	16	7.5	0.6	5.8
Rhizosphere	*Persicaria capitata*	307	958	0.6	11	3.6	5.6
Rhizosphere	*Persicaria capitata*	6.3	221	20	7.9	0.2	3
Rhizosphere	*Persicaria puncata*	20	530	8.6	2.7	0.8	18
Rhizosphere	*Conyza cordata*	13	685	15	2.8	0.3	4.2

all values in mg kg^{-1}

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