

Examination of *Blepharis aspera* as a possible Cu–Ni indicator plant

Bonang B.M. Nkoane^{a,*}, Grethe Wibetoe^b, Walter Lund^b and Berhanu Abegaz^a

BLEPHARIS ASPERA WAS COLLECTED FROM A copper–nickel mineralized area in Botswana and examined as a possible Cu–Ni indicator plant for biogeochemical prospecting. Different plant parts and the host soils were analysed using ultrasonic slurry sampling electrothermal atomic absorption spectrometry. All plant parts accumulated Cu and Ni in above-normal amounts, although not in hyperaccumulator concentrations. The leaf to soil Cu concentration ratio varied little with metal concentration in the soil. We propose *Blepharis aspera* as a Cu indicator plant.

Introduction

We present the results of a study of *Blepharis aspera* Obermeyer (family Acanthaceae), which has not been reported in Botswana before, as a possible Cu–Ni indicator plant. Recently,¹ a similar Cu–Ni study as carried out on two metal-tolerant plants (metallophytes)—*Blepharis diversispina* (Nees) C.B. Clarke (Acanthaceae) and *Helichrysum candolleianum* H. Buek (Asteraceae)—in which the latter was reported to be a possible Cu–Ni indicator. Metallophytes cope differently with high levels of metals in the soil, and therefore it is desirable to establish the metal tolerance of these plants. There are various types of metal-tolerant behaviour (displaying exclusion, accumulation and indicator) of plants that grow in mineralized soils.^{1–4}

^aDepartment of Chemistry, University of Botswana, Private Bag UB 0074, Gaborone, Botswana

^bDepartment of Chemistry, University of Oslo, P.O. Box 1033, N-0315 Oslo, Norway.

*Author for correspondence.
E-mail: bonangn@kjemi.uio.no and nkoanebbm@hotmail.com

Experimental

Blepharis aspera is a shrub with purple-bluish flowers (Fig. 1). It was identified by Kaj Vollesen, Royal Botanic Gardens, Kew, U.K. (voucher number 03SEL BD1). It grows in western Zimbabwe and northern parts of South Africa.^{5,6} Some of the plants and the host soils were collected from Selkirk, an active copper–nickel mine, in northeastern Botswana, on three sampling trips carried out on 13/14 November 2002, 12/13 March 2003 and 20/21 August 2004. The samples were air-dried and ground to a fine powder (less than 63 μm).

Copper and nickel were determined using a Perkin-Elmer Zeeman atomic absorption spectrometer (AAnalyst 800, PE; Überlingen, Germany), set at 222.6 nm for Cu and 232.0 nm for Ni, and ultrasonic slurry sampling ETAAS with nitric acid and

Triton X-100 diluent. Certified reference materials were used for method validation. Details of the analytical procedure were described previously.¹

Results and discussion

Of the four sampling locations visited in Botswana (Selkirk, Thakadu, Malaka and Nakalakwana), *B. aspera* was found only at Selkirk, which had high soil Cu and Ni concentrations in the range 0.38–1.9% Cu and 0.17–0.37% Ni. Thus, the plant can be classified as a metallophyte.⁷ To investigate if it could be used as a Cu–Ni indicator plant, it was important to know if *B. aspera* accumulated Cu and Ni in high amounts, and also to establish the type of metal tolerance (exclusion or accumulation).

Figures 2A and B show the metal concentrations in the roots, stem, leaves and flowers, for Cu and Ni, respectively, arranged according to increasing soil metal content. The plant accumulated both metals mostly in the stem or roots, and least in the flowers. The metal concentrations recorded were: 250–830 $\mu\text{g g}^{-1}$ Cu and 180–250 $\mu\text{g g}^{-1}$ Ni in the roots; 240–570 $\mu\text{g g}^{-1}$ Cu and 140–950 $\mu\text{g g}^{-1}$ Ni in the stem; 50–200 $\mu\text{g g}^{-1}$ Cu and 96–500 $\mu\text{g g}^{-1}$ Ni in



Fig. 1. Photograph of *Blepharis aspera* growing in its natural environment at Selkirk, Botswana.

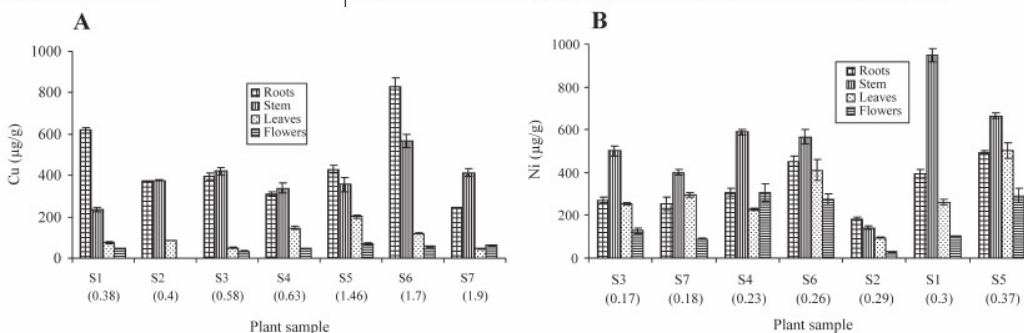


Fig. 2. Graphs of (A) Cu and (B) Ni concentrations in plant parts of *Blepharis aspera*, arranged according to increasing soil metal concentration from left to right. The soil metal concentrations (in %) are shown in brackets below the sample designation. S represents the sampling location, Selkirk (the number that follows is arbitrary). The error bars represent the standard deviation ($n = 5$). Note: the S2 flower was not available for Cu determination.

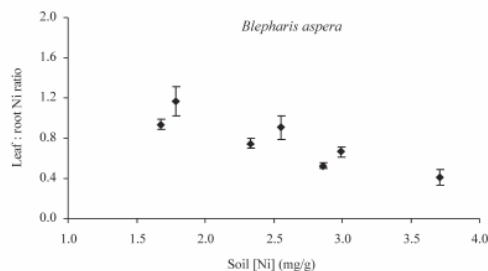


Fig. 3. Plot of leaf to root Ni concentration ratio against the metal concentration in the soil. The error bars represent the standard deviation ($n = 5$).

the leaves; and $30\text{--}70 \mu\text{g g}^{-1}$ Cu and $30\text{--}306 \mu\text{g g}^{-1}$ Ni in the flowers. These values are higher than the corresponding concentrations typically found in plants.^{8–10}

The ratio of copper concentration in leaf and root (L/R ratio) varied between 0.38% and 1.9% (mg g^{-1}) and seemed to be independent of metal concentration in the soil, whereas the Ni L/R ratio decreased with increasing metal content in the soil (Fig. 3). *Blepharis aspera* therefore behaved as a copper excluder in all soils investigated ($L/R < 1$), and as a nickel excluder in relatively high-Ni soils (concentrations greater than about 2.5 mg g^{-1}).²

The metal transfer coefficient (TC),^{8,11} which is defined as the ratio of the metal concentration in the plant to the total metal concentration in the soil, was used to estimate metal uptake and transfer to the shoot. In our study, the metal concentration in the leaf was used as an estimate of the plant concentration. The TCs for *B. aspera* were in the range 0.01–0.02 for Cu and 0.1–0.2 for Ni. The Ni TCs were higher than the corresponding copper values by an order of magnitude, indicating that for the same metal concentration in the soil, Ni had the higher uptake and translocation. The Cu TCs varied little with soil metal concentrations in the range 0.38–1.9%, suggesting a metal uptake and translocation that was proportional to the soil metal concentration. Also, the Ni

TCs did not vary much, but more data are needed over a wider range of soil Ni concentrations, to establish this plant's nickel uptake and translocation. Although the L/R ratio suggests that *B. aspera* excludes copper from the leaves, the relatively constant TCs observed for Cu imply a metal uptake and translocation proportional to the soil metal concentration. As such, *B. aspera* can be proposed as a Cu indicator plant for biogeochemical prospecting.

Our work so far has revealed that the two metallophytes, *B. aspera* and *H. candolleianum*, which grow in the same mineralized area, accumulate Cu and Ni differently in their various plant parts, implying different metal tolerances. *Helichrysum candolleianum* has a tendency to take up metals and transfer them to the higher parts (leaves and flowers), which indicates accumulator plant behaviour,¹ whereas *B. aspera* tends to restrict most of the metal to the stem and roots, a sign of excluder behaviour. Although *B. aspera* may thus be classified as an excluder plant, it still transfers some metal to the leaves in proportion to the soil metal content, especially for Cu. This suggests that it may be used for biogeochemical prospecting, in which a constant plant:soil ratio is of particular interest² because it reflects the degree of mineralization in the soil. *Helichrysum candolleianum* also

had relatively constant Cu and Ni TCs over a wide range of soil metal concentrations, implying metal uptake and translocation proportional to the soil concentration, and hence could act as a Cu–Ni indicator plant.¹ Of the two species, *H. candolleianum* may be a better Cu and Ni indicator for biogeochemical prospecting because of its ability to shift metals to its above-ground parts and thus can be more readily sampled in the field.⁷

We are grateful for financial assistance from the Norwegian Universities Committee for Development Research and Education and for the contribution from the departments of Biology (Botany) and of Chemistry, University of Botswana, in particular the project coordinator Nelson Torto. We are grateful to Kaj Vollesen, of The Royal Botanic Gardens, Kew, for the identification of *B. aspera*.

1. Nkoane B.B.M., Sawula G.M., Wibetoe G. and Lund W. (2005). Identification of Cu and Ni indicator plants from mineralised locations in Botswana. *J. Geochem. Explor.* 86, 130–142.
2. Baker A.J.M. (1981). Accumulators and excluders – strategies in the response of plants to heavy metals. *J. Plant Nutr.* 3, 643–654.
3. Baker A.J.M. and Brooks R.R. (1989). Terrestrial higher plants which accumulate metallic elements – a review of their distribution, ecology and phytochemistry. *Biorecovery* 1, 81–126.
4. Tilstone G.H. and Mcnair M.R. (1997). The consequence of selection for copper tolerance on the uptake and accumulation of copper in *Mimulus guttatus*. *Ann. Bot.* 80, 747–751.
5. Pollard A.J., Powell K.D., Harper E. and Smith J.C.A. (2002). The genetics basis of metal hyper-accumulation in plants. *Crit. Rev. Plant Sci.* 21(6), 539–566.
6. Van Wyk B. and Malan S. (1988). *Field Guide to the Wild Flowers of the Witwatersrand and Pretoria Region*. Struik, Cape Town.
7. The Flora of Zimbabwe: Online: <http://www.zimbabweflora.co.zw>
8. Adriano D.C. (2001). *Trace Elements in Terrestrial Environments: Biochemistry, Bioavailability and Risks of Metals*, 2nd edn. Springer-Verlag, New York.
9. Kabata-Pendias A. and Pendias H. (1992). *Trace Metals in Soils and Plants*, 2nd edn. CRC Press, Boca Raton, Florida.
10. Loneragan J.F. and Robson A.D. (eds), (1981). *Copper in Soils and Plants*. Academic Press, New York.
11. Henning B.J., Snyman H.G. and Aveling T.A.S. (2001). Plant–soil interactions of sludge-borne heavy metals and the effect on maize (*Zea mays* L.) seedling growth. *Water SA* 27, 71–78.