Assessing metal hyperaccumulation using herbarium specimens: the case of *Odontarrhena sibirica* (Brassicaceae)

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Metal hyperaccumulators are unusual plants that possess the rare ability to accumulate particular metals or metalloids in their living tissues to levels that may be hundreds or thousands of times greater than is normal for most species (1, 2). These plants are of substantial interest for research as they represent models to investigate the mechanisms of metal uptake, transport and sequestration, as well as adaptation and evolution in extreme environments. An active field of enquiry is also the ecology of hyperaccumulators, especially anti-herbivore defense, allelopathy and biotic interactions. These plants are also of remarkable practical interests for biotechnological applications such as phytoremediation and agromining (3). At present ca. 700 species of Angiosperms are known to be hyperaccumulators for metals such as Cadmium, Cobalt, Lead, Zinc, and Nickel. Most of them are restricted to outcrops of ultramafic rocks that are naturally rich in some of these heavy metals.

Timely identification of hyperaccumulator species is essential to promote research and applications, as well as to preserve their diversity in a rapidly changing world (4). Herbarium specimens provide a unique resource to discover hyperaccumulator species by means of several techniques, such as Atomic Absorption Spectrophotometry (AAS), inductively-coupled plasma atomic emission spectroscopy (ICP-AES), and inductively-coupled plasma mass spectrometry (ICP-MS).More recently, new analytical technology such as X-ray fluorescence (XRF) has enabled mass analysis of herbarium collections at a rate of *c.* 300 specimens per day, in a non-destructive way. Hence, new substantial discoveries are expected in the next decades by systematic screening of existing collections in herbaria around the world.

In this contribution we investigated Nickel accumulation in a facultative serpentine species from the E Mediterranean and W Asia, *Odontarrhena sibirica* (Willd.) Španiel et al. (≡ *Alyssum sibiricum* Willd.). *Odontarrhena* is the most diverse genus of Ni accumulators in W Eurasia, with some 80 species most of which grow obligatedly or facultatively on ultramafic soils and accumulate well above 1000 mg/kg of Ni in their shoots. At present, *O. sibirica* is the only species in the genus that is not clearly able of accumulating Ni even when growing on Ni-rich ultramafic soils. Previous reports for this species gave contrasting results, some of them indicating shoot Ni levels < 1000 mg/kg even in plants from ultramafic soils (Global Hyperaccumulator Database; http://hyperaccumulators.smi.uq.edu.au/collection/). We applied AAS to assess Ni content in shoots collected from multiple herbarium samples from Greece, Turkey and other countries, conserved in major European herbaria such as B, FI, G, K, and W. Based on collections details reported on the label we could assess whether the specimens originated from serpentine or non-serpentine soils, and compare Ni-levels in the two groups of edaphic accessions. Results support that *O. sibirica* may actually be the only species in its genus that is unable to accumulate Ni even when growing on serpentine soils. This might suggest an evolutionary loss of accumulation capacity, which would make of this species an ideal model to investigate the genetic bases, physiological mechanisms and molecular pathways of this unique ability.

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