Supporting Information

to

Breeding the hyperaccumulator *Noccaea caerulescens* for trace metal phytoextraction: First results of a pure-line selection

Thibault Sterckeman, Yannick Cazes, Catherine Sirguey

Université de Lorraine, Inra, Laboratoire Sols et Environnement, F-54000 Nancy, France

Corresponding author: Thibault Sterckeman

E-mail: Thibault.Sterckeman@univ-lorraine.fr
Table S1. Location and code of 60 populations of *Noccaea caerulescens* used for the experiment. EG: Edaphic group; CAL: Calamine; NM: Non Metalliferous; SER: Serpentine.

| Edaphic group | Code | Location         | Europe | Country               | Coordinate         |
|---------------|------|------------------|--------|-----------------------|--------------------|
| CAL           | B01  | Prayon           | West   | Belgium               | 50°34'38.05"N 05°40'28.02"E |
| CAL           | B03  | La Calamine      | West   | Belgium               | 50°41'44.02"N 05°59'39.40"E |
| CAL           | F03  | Auxelles         | West   | France                | 47°44'27.96"N 06°46'31.63"E |
| CAL           | F05  | Auxy             | West   | France                | 46°57'44.13"N 04°23'47.05"E |
| CAL           | F19  | La Galène        | West   | France                | 42°56'48.10"N 00°05'11.95"W |
| CAL           | F44  | Largentière      | West   | France                | 44°32'26.33"N 04°18'18.44"E |
| CAL           | F45  | Ste Marguerite   | West   | France                | 44°27'29.13"N 04°00'31.32"E |
| CAL           | F46  | Le Bleyvard      | West   | France                | 44°28'18.52"N 03°43'40.45"E |
| CAL           | F47  | Ramponenche      | West   | France                | 44°20'15.85"N 03°40'00.31"E |
| CAL           | F48  | St Félix         | West   | France                | 44°02'36.21"N 03°56'11.27"E |
| CAL           | F49  | Durfort           | West   | France                | 43°59'56.70"N 03°57'08.54"E |
| CAL           | F50  | Malines          | West   | France                | 43°55'20.67"N 03°37'14.57"E |
| CAL           | F51  | Sanguinède       | West   | France                | 43°55'56.80"N 03°36'29.12"E |
| CAL           | F52  | Ganges           | West   | France                | 43°56'10.98"N 03°40'19.88"E |
| CAL           | F54  | Viviez           | West   | France                | 44°33'34.16"N 02°13'01.39"E |
| CAL           | F64  | Villemagne       | West   | France                | 44°07'19.98"N 03°26'16.87"E |
| CAL           | G1   | Hasbergen        | West   | Germany               | 52°13'48.83"S 07°56'34.91"E |
| CAL           | SP2  | Pontaut          | South  | Spain                 | 42°50'40.57"N 00°43'56.07"E |
| CAL           | SP4  | Lanestosa         | South  | Spain                 | 43°12'56.64"N 03°26'14.39"W |
| CAL          | SLK01 | Térélia mine     | East   | Slovakia              | 48°26'21.00"N 18°52'12.00"E |
| NM           | F10  | Montmelas-Saint-Sorlin | West | France            | 46°01'14.40"N 04°35'27.54"E |
| NM           | F13  | Pierre-Pertuis    | West   | France                | 47°26'12.63"N 03°46'59.42"E |
| NM           | F27  | Pic de Chenavari  | West   | France                | 44°35'58.80"N 04°41'04.20"E |
| NM           | F29  | Ste-Eulalie      | West   | France                | 44°48'13.10"N 04°12'47.10"E |
| NM           | F32  | L’Herm           | West   | France                | 44°54'24.20"N 03°49'10.10"E |
| NM           | F34  | Montchamp        | West   | France                | 45°04'06.80"N 03°12'45.40"E |
| NM           | F38  | La Chavignéée    | West   | France                | 45°23'48.30"N 02°43'59.00"E |
| NM           | F40  | Besse            | West   | France                | 45°30'25.50"N 02°56'26.30"E |
| NM           | F43  | Col des Supeyres | West   | France                | 45°35'24.10"N 03°50'55.20"E |
| NM           | F53  | Laguiole         | West   | France                | 44°40'50.30"N 02°55'51.54"E |
| NM           | F61  | St-Maurice       | West   | France                | 43°53'16.61"N 03°30'29.33"E |
| NM           | F66  | Le Cernois       | West   | France                | 46°39'11.81"N 06°07'59.79"E |
| NM           | F67  | Prémonan         | West   | France                | 46°27'38.18"N 06°00'50.25"E |
| NM           | F68  | Sepmoncel        | West   | France                | 46°22'31.05"N 05°54'27.66"E |
| NM           | F71  | Fresse sur Moselle | West | France            | 47°52'18.50"N 06°47'29.50"E |
| NM           | F72  | Geishouse        | West   | France                | 47°53'32.50"N 07°04'19.60"E |
| NM           | F74  | Cornimont        | West   | France                | 47°59'03.60"N 06°49'05.30"E |
| NM           | F75  | Col d’Oderen     | West   | France                | 47°54'50.70"N 06°55'11.40"E |
| NM           | F78  | Buèges           | West   | France                | 43°49'47.31"N 03°37'06.74"E |
| NM           | F79  | Baraquette       | West   | France                | 43°55'07.04"N 03°36'56.57"E |
| NM           | F80  | Mas St Chely     | West   | France                | 44°18'33.79"N 03°23'46.16"E |
| Code | Location                  | Direction | Country      | Coordinates                  |
|------|---------------------------|-----------|--------------|------------------------------|
| NM   | Pornainen                 | North     | Finland      | 60°28'30.21"N 25°22'19.65"E  |
| NM   | Großschönau               | West      | Germany      | 50°50'56.71"N 14°38'48.59"E  |
| NM   | Dresde                    | West      | Germany      | 51° 4'29.34"N 13°45'42.54"E  |
| NM   | Wilwerwiltz               | West      | Luxembourg   | 49°59'28.52"N 05°59'39.68"E  |
| NM   | Winseler                  | West      | Luxembourg   | 49°57'58.38"N 05°53'22.27"E  |
| NM   | Lellingen                 | West      | Luxembourg   | 49°59'07.55"N 06°00'55.80"E  |
| NM   | Vel'ká Fatra Mountain     | East      | Slovakia     | 48°36'14.00"N 18°42'24.87"E  |
| NM   | Valle de Varrados          | South     | Spain        | 42°47'05.97"N 00°41'31.51"E  |
| NM   | Uppsala                   | North     | Sweden       | 59°50'06.06"N 17°36'7.65"E   |
| NM   | Fäberg                    | North     | Norway       | 61° 9'53.35"N 10°21'44.66"E  |
| NM   | Col de Maraichuz           | West      | Switzerland  | 46°32'26.84"N 06°15'20.94"E  |
| NM   | Chasseron                 | West      | Switzerland  | 46°52'04.81"N 06°33'17.11"E  |
| NM   | Mont d'Amin               | West      | Switzerland  | 47°05'11.61"N 06°54'06.70"E  |
| NM   | Tête de Ran               | West      | Switzerland  | 47°03'20.29"N 06°51'23.19"E  |
| NM   | Ulrichen                  | West      | Switzerland  | 46°30'14.11"N 08°19'11.68"E  |
| SER  | Fellering                 | West      | France       | 47°54'13.60"N 06°57'12.07"E  |
| SER  | Puy de Wolf               | West      | France       | 44°33'23.80"N 02°18'21.07"E  |
| SER  | Montgenèvre               | West      | France       | 44°54'20.85"N 06°43'32.96"E  |
| SER  | Monte Prinzeria           | West      | Italy        | 44°38'37.07"N 10° 4'55.89"E  |
Table S2. Characteristics of the soil used for the cultivation of *Noccaea caerulescens*.

| Characteristic          | Value       | Unit       |
|-------------------------|-------------|------------|
| Clay                    | 100         | g kg⁻¹     |
| Silt                    | 314         | g kg⁻¹     |
| Sand                    | 586         | g kg⁻¹     |
| pH                      | 7.34        |            |
| CaCO₃                   | < 1         | g kg⁻¹     |
| Organic C               | 12.1        | g kg⁻¹     |
| Organic N               | 1.03        | g kg⁻¹     |
| C/N                     | 11.7        |            |
| CEC                     | 7.75        | cmol+ kg⁻¹ |
| Exchangeable Ca         | 7.72        | cmol+ kg⁻¹ |
| Exchangeable K          | 0.249       | cmol+ kg⁻¹ |
| Exchangeable Mg         | 0.23        | cmol+ kg⁻¹ |
| P₂O₅ Olsen              | 0.126       | g kg⁻¹     |
| Total Cd total          | 0.12/8.6*   | mg kg⁻¹    |
| Total Cu                | 10.7/40.2*  | mg kg⁻¹    |
| Total Ni                | 11.2/196*   | mg kg⁻¹    |
| Total Pb                | 38.8/74.1*  | mg kg⁻¹    |
| Total Zn                | 50.3/993*   | mg kg⁻¹    |

* Initial content/Content after spiking
Table S3. Selection criteria of the plants. Cd, Ni and Zn refer to the respective metal content in the rosette at the first bud visible stage (FBV). SB is the shoot biomass at final harvest. The plants of each generation were ranked according to each of the criteria and the top individuals were selected for their progeny to be cultivated the following year.

| Edaphic group | Code of SPF | SPF1 | SPF2 | SPF3 |
|---------------|-------------|------|------|------|
| CAL           | L20 B1      | Cd   | SB   | SB   |
| CAL           | L20 D3      | Cd   |      |      |
| CAL           | L21 A6      | Cd   | Cd, SB| Cd, SB|
| CAL           | L21 A6      | Cd   | Cd, SB| Cd, SB|
| CAL           | L22 D5      | Cd   | Cd   | Cd, SB|
| CAL           | L22 D5      | Cd   | Cd   | Cd   |
| CAL           | L22 D5      | Cd   | Cd   | Cd   |
| CAL           | L22 D5      | Cd   | Cd   | Cd, SB|
| CAL           | L22 D6      | Cd   | Cd   | Cd, SB|
| CAL           | L22 D6      | Cd   | Cd   | Cd   |
| CAL           | L22 D6      | Cd, SB| Cd, SB| Cd, SB|
| NM            | L05 C5      | Ni, Zn|      |      |
| NM            | L18 B4      | Ni, Zn| Ni, Zn| Ni, Zn|
| NM            | L18 B4      | Ni, Zn| Ni, Zn| Ni, Zn|
| NM            | L18 B4      | Ni, Zn| Ni, Zn| Ni, Zn|
| NM            | L18 B4      | Ni, Zn| Ni, Zn| SB, Ni|
| NM            | L18 B4      | Ni, Zn| Ni, Zn| Ni, Zn|
| NM            | L18 B4      | Ni, Zn| Ni, Zn| Ni, Zn|
| NM            | L16 E6      | SB   |      |      |
| CAL           | L23 E5      | SB   | SB   | SB   |
| CAL           | L23 E5      | SB   | SB   | SB   |
| CAL           | L23 E5      | SB   | SB   | SB   |
| CAL           | L23 E5      | SB   | SB   | SB   |
| CAL           | L15 C3      | Zn   | Zn   | Zn   |
| NM            | L15 C3      | Zn   | Ni, Zn| Ni, Zn|
| NM            | L17 E4      | Zn   | Zn   | Zn   |
| NM            | L17 E4      | Zn   | Zn   | Ni, Zn|
| NM            | L17 E4      | Zn   | Zn   | Zn   |
| NM            | L17 E4      | Zn   | Zn   | Cd, Ni|
| NM            | L17 E4      | Zn   | Zn   | Zn   |
| NM            | L16 A4      | Zn, SB| Zn| Zn|
Figure S1. Location of the 60 populations of *Noccaea caerulescens* sampled in Europe, according to three edaphic groups. NM: Non-metallicolous; CAL: Calamine; SER: Serpentine.
Figure S2. Plan and picture of the cultivation device.
Figure S3. Cumulated thermal time of the three years of cultivation.

Figure S4. Relationship between the shoot dry matter of the plants selected in F1 (SPF1) and that of their progenies in F2.
Figure S5. Relationship between the shoot dry matter of the plants selected in F2 (SPF2) and that of their progenies in F3.
Figure S6. Relationship between the rosette Cd content of a) the plants selected in F1 (SPF1) and that of their progenies in F2 and of b) the plants selected in F2 (SPF2) and that of their progenies in F3.
Figure S7. Relationship between Cd content in rosette at FBV and in shoot at FRR for the three cultivation years.
Figure S8. Relationship between sowing density of the different generations (F1, F2 and F3) and the mean Cd offtake.

Figure S9. Cadmium offtake of the selected plants of each generation.
Figure S10. Relationship between the rosette Ni content of a) the plants selected in F1 (SPF1) and that of their progenies in F2 and of b) the plants selected in F2 (SPF2) and that of their progenies in F3.
Figure S11. Relationship between Ni content in rosette at FBV and in shoot at FRR for the three cultivation years.
Figure S12. Nickel offtake of the plants selected from each generation.
Figure S13. Relationship between the rosette Zn content of a) the plants selected in F1 (SPF1) and that of their progenies in F2 and of b) the plants selected in F2 (SPF2) and that of their progenies in F3.
Figure S14. Relationship between Ni content in rosette at FBV and in shoot at FRR for the three cultivation years.
Figure S15. Zinc offtake of the plants selected from each generation.