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# INVESTIGATIONS ON A NICKEL ACCUMULATING PLANT: ALYSSUM BERTOLONII DESV. II. PHOSPHORUS, POTASSIUM, IRON AND TRACE ELEMENT CONTENT AND DISTRIBUTION DURING GROWTH

**Abstract** — The distribution of phosphorus, potassium and iron and of some trace elements (manganese, zinc, copper, cobalt and chromium) in the growth cycle of *Alyssum bertolonii*, collected on two serpentine outcrops, Monte Murlo (Arezzo) and Impruneta (Florence), is here reported.

In addition to plant analysis, data on the soil composition are also given.

The plant phosphorus content shows that this element is often at deficiency level; rather low are also manganese and copper concentrations.

Seasonal variation in cobalt content closely corresponds to that of nickel; between these elements there is also a highly significant correlation ($P > 0.001$).

**Riassunto** — *Ricerche su una pianta accumulatrice di nichel:* Alyssum bertolonii *Desv. II. Contenuto in fosforo, potassio, ferro e oligoelementi e loro distribuzione durante il ciclo vegetativo.* E' riportata in questa nota la distribuzione di alcuni elementi macronutritivi e di diversi oligoelementi (manganese, zinco, rame, cobalto e cromo) durante un intero ciclo vegetativo di *Alyssum bertolonii,* raccolto sugli affioramenti ofiolitici di Monte Murlo (Arezzo) e di Impruneta (Firenze). In una nota precedente (WEBBIA 1977, 32: 175) erano state prese in considerazione le variazioni stagionali nel contenuto in nichel, calcio e magnesio.

I risultati mostrano che il contenuto in fosforo, sopra tutto nei campioni provenienti da Monte Murlo, è molto scarso e talvolta a livelli carenziali. Piuttosto basso il contenuto in manganese e rame. L'andamento stagionale del cobalto corrisponde notevolmente a quello del nichel; tra i due elementi esiste anche una correlazione altamente significativa ($P > 0.001$).

**Key words** — *Alyssum bertolonii* - element content and distribution.

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In the previous paper (VERGNANO GAMBI et al., 1977) calcium, magnesium and nickel distribution was reported for *Alyssum bertolonii* Desv., collected during a whole year on two Tuscan serpentine outcrops, namely Monte Murlo (Arezzo) and Impruneta (Florence). In order to give more detailed and complete information on the mineral composition of this plant we present here the results of the analyses for phosphorus, potassium, iron and other trace elements, carried out on the same samples.

## METHODS

Soil samples were air dried and passed through a 2 mm sieve. Extraction with 2.5% acetic acid was carried out by shaking 5 g soil for 24 h with 50 ml of solution. On the filtered extracts potassium, iron and trace elements were determined by atomic absorption spectrophotometry. Nitrates and phosphates were determined colorimetrically.

The results are shown in table 1.

TAB. 1

| | Total % | | Acetic acid sol. µg/g | |
|---|---|---|---|---|
| | M.Murlo | Impruneta | M.Murlo | Impruneta |
| $NO_3$ | 0.011 | 0.019 | | |
| P | 0.005 | 0.023 | 3 | 6 |
| K | 0.06 | 0.25 | 34 | 34 |
| Ca | 1.10 | 0.81 | | |
| Mg | 20.30 | 14.25 | | |
| Fe | 5.3 | 9.5 | 7 | 7 |
| Mn | 0.11 | 0.20 | 78 | 87 |
| Cu | 0.003 | 0.0045 | 0.45 | 0.50 |
| Zn | 0.007 | 0.021 | 1.4 | 1 |
| Ni | 0.20 | 0.25 | 88 | 88 |
| Co | 0.021 | 0.031 | 5.8 | 11.1 |
| Cr | 0.085 | 0.48 | 2.9 | 2.2 |

On the same plant material as used for previous analyses (VERGNANO GAMBI et al., 1977, p. 177) potassium, iron and trace elements were analysed by atomic absorption spectrophotometry. Phosphorus was determined colorimetrically as molybdenum blue, and on some samples total nitrogen was assessed by a semimicro Kjeldahl method (BAKER, 1961).

The results are reported in table 2 and in figures 1, 2 and 3.

## RESULTS AND DISCUSSION

### *Soil analysis*

In both soils, which can be described as skeletal (« sols squelettiques ») or lithosolic, organic matter is almost absent, the sand fraction (< 2 mm) reaching about 20%. *Nitrate* content is very low, particulary at M. Murlo where the average is below the lowest normal level.

Another typical feature of serpentine soils is the low *phosphorus* level (DUVIGNEAUD, 1966, PROCTOR and WOODELL, 1971, LEE et al., 1977); this is also a characteristic of these soils, particularly at Monte Murlo (0.005%) where the phosphorus content is close to that reported by KRAUSE (1958) as typical for serpentines (0.004%).

Also the *potassium* content, often at deficiency levels in serpentine soils, at M. Murlo shows typical values for these soils.

The amount of *calcium* found in the soil is generally higher than that found in the rock, probably due to weathering of nearby formations, richer in calcium. On the other hand the *magnesium* content shows a tendency to decrease in the soil. Typical of serpentine soils is the content at M. Murlo (~ 20%).

In serpentine soils *iron* reaches high total values (LEE et al., 1977, SLINGSBY and BROWN, 1977), although it may be only slightly soluble. Analytical data on this element vary greatly but in our samples the iron values are higher at Impruneta (9.5%), and more typical of serpentines (5.8%) at M. Murlo. Acetic acid extraction confirms the restricted availability of this element.

*Manganese* values ranging between 0.08% and 0.46% have been recorded for serpentine soils of Great Britain (SLINGSBY and BROWN, 1977) and higher contents (0.6%) have been observed (LEE et al., 1977) in New Caledonia, but generally the manganese content of serpentines is low, 0.18% being the average according to KRAUSE (1958), so that manganese deficiency is not infrequent. At M. Murlo and Impruneta the total content ranges between 0.11 and 0.20%; acetic acid extraction gives similar values for the two localities (78 and 87 μg/g). Because its content in serpentine is not high, *zinc* is seldom considered in geochemical analyses; at M. Murlo total zinc reaches 70 μg/g and at Impruneta 210 μg/g, but acetic acid extraction does not show appreciable differences between the two areas. Higher total values (193 - 315 μg/g) have been observed by LEE et al. (1977) in samples of serpentine soils from New Caledonia, where the nickel accumulating plants have higher zinc contents.

The total *copper* content of the soil is quite low (30-45 µg/g), as is often the case in serpentine soils (WILD, 1974 b, PROCTOR and JOHNSTON, 1977). Higher values (58-82 µg/g) have been observed for this element by LEE et al. (1977) in New Caledonia. The extractable fraction (0.45 - 0.50 µg/g) is very low, as observed also by WILD (1974 b).

The total *cobalt* content (210 - 315 µg/g) of our samples corresponds to that most frequently observed in serpentine soils (ERNST, 1972, LYON et al., 1970, SLINGSBY and BROWN, 1977). Higher values have been found in Japan (in PROCTOR and WOODELL, 1975) and in New Caledonia (800 µg/g) by LEE et al. (1977). In all samples collected at M. Murlo the acetic acid soluble cobalt is similar, with rather high values ($\sim$ 6 µg/g): at Impruneta this fraction shows quite variable values (18.8 - 3.2 µg/g) depending on the organic matter content of the soil.

The total *chromium* content is notably different in the two localities: the average content is 0.48% in the Impruneta soil, almost double the content found in the rock (0.27%) and higher than the average chromium content of serpentines (KRAUSE, 1958). It is only 0.085% at M. Murlo, where it closely correponds to the total chromium content (0.08%) of the ophiolitic breccia (BRUNACCI et al., 1976). Values found at Impruneta are similar to those observed by LYON et al. (1970) in New Zealand and by LEE et al. (1977) in New Caledonia; the chromium content at M. Murlo is closer to the values found by WILD (1974 a). The acetic acid soluble fraction is almost the same in both localities: 2.9 µg/g at Monte Murlo, 2.2. µg/g at Impruneta.

We can conclude this brief survey on the soil composition by noting that M. Murlo reflects well the typical characters of a serpentine soil representing the first weathering stages of the parent rock and therefore showing a close relationship with the rock composition: magnesium, calcium and chromium have almost the same concentration as in the parent rock, only iron and manganese reaching slightly higher values.

At Impruneta the underlying rock is covered with a brown-reddish soil, in which notably chromium and iron have high concentrations, whilst magnesium has been partly removed. The ophiolitic outcrop being included among formations of different geological nature accounts for the increase in soil calcium content.

## *Plant analysis*

In Table 2 we have reported the average values on dry weight of all the elements considered, including nickel, calcium and magnesium listed in the previous paper (WEBBIA, 1977, p. 179). Values refer to the mean of all samples analysed excluding only the first

TAB. 2 - Mean elemental concentration, on dry weight (dw) of various organs of *Alyssum bertolonii*, collected at Monte Murlo and Impruneta.

| | N | P | K | Ca<br>% | Mg | Ni | Co | Cr | Fe<br>μg/g | Mn | Zn | Cu |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| *Monte Murlo* | | | | | | | | | | | | |
| leaves: | | | | | | | | | | | | |
| green | 2.07 | 0.11 | 1.23 | 3.73 | 0.61 | 10.600 | 124 | 6 | 418 | 255 | 18 | 5 |
| purple | | 0.05 | 1.14 | 2.85 | 0.68 | 9.200 | 195 | 7 | 656 | 227 | 11 | - |
| stems: young | | 0.12 | 1.48 | 1.37 | 0.34 | 5.240 | 8 | 3 | 171 | 74 | 26 | - |
| woody | | | | 0.55 | 0.15 | 1.290 | 3 | 2 | 207 | 23 | 19 | - |
| sprouts | | 0.10 | 1.26 | 3.18 | 0.75 | 8.300 | 68 | 7 | 291 | 301 | 12 | - |
| roots | | 0.10 | 0.79 | 0.40 | 0.31 | 1.220 | 8 | 13 | 1010 | 45 | 14 | - |
| flowers | 2.90 | 0.30 | 1.96 | 1.70 | 0.56 | 7.310 | 18 | 4 | 201 | 142 | 31 | - |
| fruits | | | | 1.73 | 0.44 | 6.630 | 12 | 3 | 98 | 66 | 19 | - |
| seeds | 3.04 | 0.50 | 2.77 | 0.75 | 0.19 | 7.370 | 19 | 5 | 66 | 105 | 30 | - |
| *Impruneta* | | | | | | | | | | | | |
| leaves: | | | | | | | | | | | | |
| green | 3.33 | 0.17 | 1.75 | 3.44 | 0.69 | 9.260 | 73 | 8 | 467 | 140. | 49 | 10 |
| stems: young | | 0.16 | 1.61 | 1.59 | 0.39 | 4.480 | 6 | 3 | 191 | 42 | 49 | - |
| Woody | | | | 0.48 | 0.18 | 2.220 | 3 | 5 | 408 | 24 | 63 | - |
| roots | | 0.05 | 0.75 | 0.51 | 0.36 | 2.050 | 11 | 41 | 1710 | 74 | 48 | - |
| flowers | 3.13 | 0.27 | 2.19 | 1.98 | 0.52 | 6.860 | 10 | 5 | 172 | 115 | 66 | - |
| fruits | | | | 0.97 | 0.40 | 4.910 | 13 | 2 | 87 | 46 | 31 | - |
| seeds | 3.47 | 0.40 | 3.40 | 0.87 | 0.19 | 4.390 | 9 | 2 | 58 | 41 | 48 | - |

M. Murlo sampling (early February 1975) which represents a rather atypical condition. In figs. 1-3 the amount of the elements, from February-March to October-November at M. Murlo and Impruneta, are illustrated.

## *Nitrogen*

Nitrogen values reported by DUVIGNEAUD (1966) for some serpentine species growing in S-W France, range between 0.7 and 3.1%. The nitrogen content of *A. bertolonii* (Tab. 2) is therefore rather high, particularly at Impruneta where the nitrates content of the soil is also higher. In plants of *A. bertolonii* grown on normal garden soil, a maximum nitrogen content of 3.6% has been observed; therefore nitrogen content can be considered normal at Impruneta and adequate at Monte Murlo, taking also in account the good development of plants from this locality.

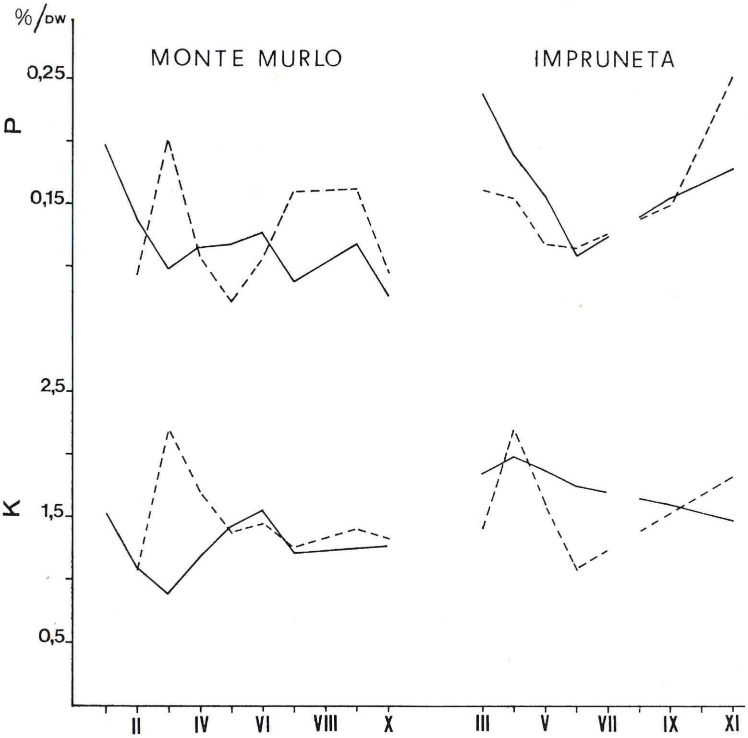


Fig. 1 - Seasonal variation in phosphorus and potassium content (% d.w.) of leaves (solid line) and green stems (dotted line) of *A. bertolonii* at M. Murlo and Impruneta.

## *Phosphorus*

At M. Murlo the leaves of *A. bertolonii* have a maximum phosphorus level of 0.20% with an average value of 0.12%, similar to that (0.10%) found by DUVIGNEAUD in the serpentine vegetation of S-W France. Lower phosphorus concentrations have been observed in two nickel accumulating plants by LEE et al. (1977) and occasionally in some M. Murlo plants — in which phosphorus was at deficiency levels (0.034-0.040%) — a typical red tinting diffusing into stems and leaves, quite different from the purple colouring of older leaves. No thing similar occurred at Impruneta, where the leaf phosphorus content is never lower than 0.10% and the soil content is also higher.

In both localities the phosphorus content is highest when the

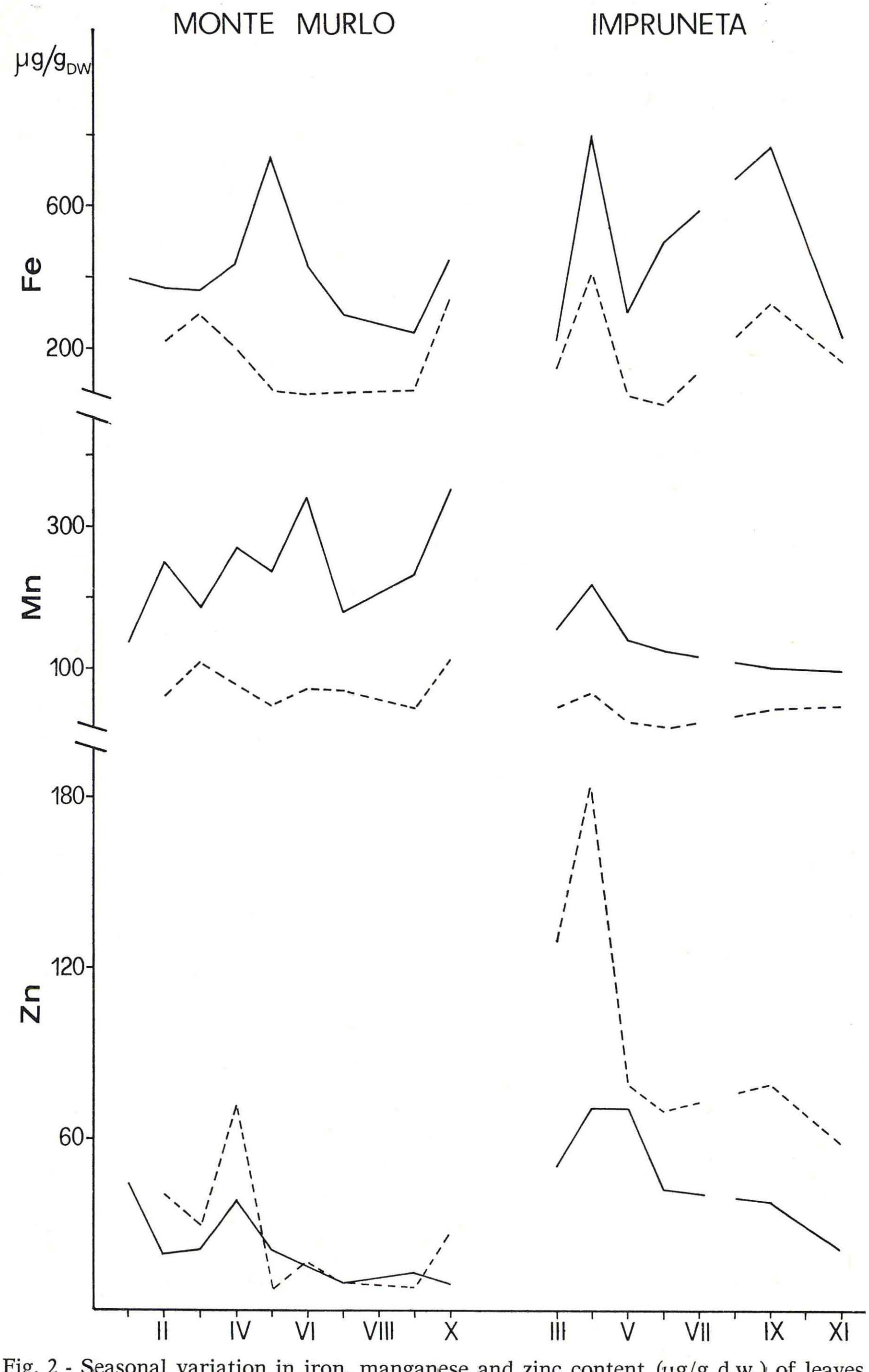


Fig. 2 - Seasonal variation in iron, manganese and zinc content (µg/g d.w.) of leaves (solid line) and green stems (dotted line) of *A. bertolonii* at M. Murlo and Impruneta.

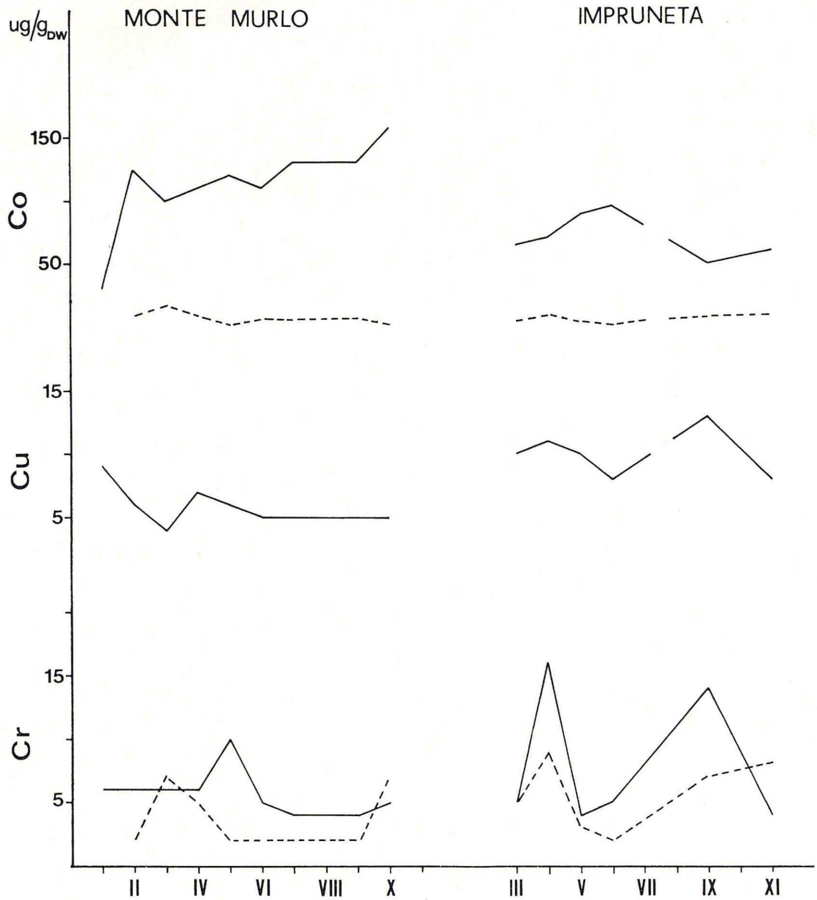


Fig. 3 - Seasonal variation in cobalt, copper and chromium content (μg/g d.w.) of leaves (solid line) and green stems (dotted line) of *A. bertolonii* at M. Murlo and Impruneta.

plants resume growth (fig. 1), the distribution pattern at Impruneta being similar in leaves and stems whereas at M. Murlo distribution is different in the two organs and sometimes opposite; the content of the green stems is highest at M. Murlo in March, and at Impruneta in April.

## *Potassium*

The potassium content (fig. 1) also, is higher in the Impruneta plants and shows slight seasonal variations (1.48--1.97% dw) as compared with the M. Murlo plants (0.88-1.55%), which have a minimum value in March, when the green stems show the highest value (2.12%). The same occurs a month later at Impruneta.

Values are lower (1.14%) in the purple leaves, particularly (0.7%) in the P-deficient leaves. There are no differences between the two localities at root level (table 2). The content in flowers and seeds is remarkable, so that, compared with others, this element may be present in relatively adequate quantities. The Impruneta soil shows higher values for total K content, 0.25%, which might explain the higher content in the plants from this locality. DUVIGNEAUD (1966) has also observed some high potassium contents in serpentine plants, whilst LEE et al. (1977) report lower values for *Hybanthus* and *Homalium*, although in all cases seasonal variations must also be taken into account.

*Iron*

The average iron leaf content (~ 400 μg/g dw) is almost the same in both localities (tab. 1) ranging between 245 and 742 μg/g at Monte Murlo and 225 and 791 μg/g at Impruneta; the older leaves reach occasional high values (290-1180 μg/g dw). The highest concentrations are in roots, particularly at Impruneta.

In the Impruneta samples, the seasonal variations (fig. 2) in leaves and green stems are very similar, with a maximum in April and another in September. At M. Murlo the leaves reach their maximum iron content in May, the stems in March.

The iron content of *A. bertolonii* is comparable with that observed by LEE et al. (1977) in *Hybanthus* and by ERNST (1972) in *Indigofera setiflora* from mining areas in Rhodesia. In this case also the highest values were observed in the roots.

*Manganese*

Notwithstanding the low Mn content of the soil, the leaf content at M. Murlo (138-352 μg/g dw; ~ 1600 μg/g a w*), and also at Impruneta (101-218 μg/g dw; ~ 900 μg/g aw), is similar to that of plants from other serpentine areas (ERNST 1972, LEE et al. 1977), and the values are within the variability observed on other substra-

(*) aw = ash weight.

tes (BEESON et al., 1955; LAZAR et al., 1956); the average ash value quoted by CANNON (1960) is much higher, as are the Mn contents observed by KELLY et al. (1975).

At M. Murlo this element has two maxima (Summer and Autumn), and the average manganese content is higher (242 μg/g dw) than at Impruneta (140 μg/g), where there is only one maximum. The seasonal variation of the green stems is slight and the content low (18-77 μg/g dw I; 45-114 μg/g dw M.M.). No accumulation occurs at root level.

*Zinc*

The zinc concentration of the leaves of *A. bertoloni* (tab. 2 and fig. 2) is rather low at M. Murlo; similar values have been observed by BEESON et al. (1955) and by ERNST (1972) without deficiency symptoms arising. The green stems, flowers and seeds, particularly at Impruneta, show the highest accumulation.

*Copper*

*A. bertolonii* shows a copper content (fig. 3) ranging from 8 to 13 μg/g dw (51-96 μg/g aw) at Impruneta and between 4 and 9 μg/g dw (25-45 μg/g aw) at M. Murlo. This range corresponds to that observed in *Hybanthus floribundus* (COLE, 1973), and in other nickel accumulating plants from Rhodesia (WILD, 1974a) and New Caledonia (LEE et al., 1977). A similar copper content has been observed in the great majority of the serpentine plants analysed by LYON et al. (1970). The generally low copper values of serpentine plants is further confirmed by the level found by DUVIGNEAUD (1963) in plants from areas with low copper (20 ppm) in the soil.

*Cobalt*

The leaf content in *A. bertolonii* (fig. 3) ranges between 51 and 95 μg/g dw (375-732 μg/g aw) at Impruneta and between 31 and 158 μg/g dw (155-1211 μg/g aw) at M. Murlo. In other nickel accumulating plants lower cobalt values have been observed in some species of the genus *Hybanthus* (COLE, 1973; KELLY et al., 1975; and LEE et al., 1977), but generally the cobalt concentrations recorded are higher as, for example, in serpentine plants from New

Zealand (Lyon et al., 1968 and 1970) and in plants from mining areas in Rhodesia (Wild, 1974a). High cobalt concentrations have also been found in *Rinorea bengalensis* (Brooks and Withers, 1977).

In *A. bertolonii* there is a positive correlation between cobalt and nickel, both being preferentially accumulated in the leaves, cobalt particularly in the old, purple samples; woody stems and roots show the lowest content. Contents in flowers, fruits and seeds are very similar. Seasonal variation in the leaf content closely resembles that of nickel: the accumulation in autumn is very evident. The green stem content shows a maximum (16 µg/g dw; 219 µg/g aw) in March at M. Murlo and in April at Impruneta (9 µg/g dw; 150 µg/g aw). The seasonal variation in the green stems is remarkably similar at both locations.

*Chromium*

The higher chromium content of serpentines, as compared with cobalt, does not indicate a greater mobility and solubility of the element, which is generally scarcely translocated in serpentine plants: in *A. bertolonii* it is mainly bound at root level (13 and 41 µg/g dw; 305 and 745 µg/g aw at Impruneta and Monte Murlo respectively). The leaf content ranges between 4 and 16 µg/g dw (25-103 µg/g aw) at Impruneta, and between 4 and 10 µg/g dw (29-62 µg/g aw) at M. Murlo. Similar concentrations have been observed in *Hybanthus floribundus* by Cole (1973) and in other nickel accumulating species by Kelly et al. (1975) and Lee et al. (1977); higher values have been observed by Wild in Rhodesia (1974a). The seasonal variation (fig. 3) in the Impruneta samples is similar to that of other trace elements.

From these analyses it is clear that although the levels of calcium and potassium can be considered sufficient, particularly at M. Murlo, values for phosphorus (and occasionally nitrogen) can be very low. In the leaf samples from both localities, significant negative relationships, on ash basis, have been observed between calcium and magnesium ($P > 0.05$), and between calcium and potassium ($P > 0.001$).

The inter-elemental relationships within the plant show a positive, highly significant ($P > 0.011$) correlation between cobalt and nickel, as observed by Lyon et al. (1968) in serpentine plants from

New Caledonia. BROOKS et al. (1977) also noticed the absence of antagonism in the uptake of these elements in plants accumulating nickel or cobalt. A positive correlation ($P > 0.001$) is shown also between cobalt and manganese.

No other significant relationship for nickel, chromium and cobalt has been found in these samples. A tendency to a regular increase in magnesium content with increasing nickel concentration points to a relationship between these elements, which reaches a significant level ($P > 0.01$) if values for stems plus leaves are considered. Positive and significant correlations between chromium and copper ($> 0.02$) and between chromium and iron ($P > 0.001$), are clearly shown in both localities.

Seasonal variation in the concentration of the elements at M. Murlo shows a close relationship between the content in stems and leaves, the maximum leaf content generally corresponding to the lowest stem value. For phosphorus and potassium the minimum leaf content (and the maximum for the stems) occurs in March; for iron, cobalt, chromium and to a lesser extent, for manganese, the maximum leaf accumulation occurs in May. The seasonal fluctuation in the stem content is practically the same for most elements, with a peak in March, the lowest content during Summer and a more or less marked increase in Autumn.

For zinc, both in stems and leaves, the pattern is quite different, a maximum occurring in April, when manganese, copper and calcium also show high values.

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