

Relationships between specific leaf area and leaf composition in succulent and non-succulent species of contrasting semi-desert communities in south-eastern Spain



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ABSTRACT

Much attention has been paid to differences in leaf form and composition among vegetation types, but less to the frequently substantial variation within vegetation types. We focused on the extent to which correlations between variables are the same in both succulent-poor and succulent-rich vegetation in semi-arid SE Spain. Mean foliar [N] of perennials varied among species over a 5-fold range. Across species, [N] was positively correlated with specific leaf area (i.e., leaf area divided by dry mass; SLA) and with water concentration at saturation (WCS) in the grasslands, excluding the one succulent species. In succulent-rich vegetation on marl, SLA was correlated with [N] but not WCS, and there was a wedge-shaped relationship between [N] and WCS. Foliar [N] and [P] were positively correlated in the grasslands, but not in succulent-rich vegetation on marl. The N/P quotient varied from 8 to 29, with mean 14 in grassland on limestone and mean 26 in grassland on deep soil over gypsum. Our chief finding is that most correlations among SLA, WCS, [N] and [P] found in the non-succulent vegetation are not found in the succulent-rich vegetation. The results are discussed in relation to global patterns and the problems of defining succulence.

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1. Introduction

Much attention has been paid to differences in leaf form and composition among vegetation types, but less to the frequently substantial variation within vegetation types. This paper provides the first substantial information on leaf properties of perennial plants of semi-desert in the only part of Europe – except possibly south-eastern Crete (Grove and Rackham, 2001) – dry enough to carry such vegetation, i.e. southern Spain between Almería and Cartagena (Armas et al., 2011, Fig. 1). We use the term ‘semi-desert’ for areas with a diffuse incomplete cover of perennials, in contrast to true ‘desert’ in which perennials may occur extremely sparsely or only at run-on sites (Walter, 1964; Bornkamm and Kehl, 1990). Most of the semi-desert of SE Spain has a mean annual rainfall of 200–250 mm yr⁻¹ with some sites receiving <190 mm yr⁻¹ (Lázaro

et al., 2001; see Appendix 1 for the long-term and recent history of the area, and the nature of its present vegetation cover). The area encompasses a variety of communities, depending on altitude, aspect, soil type and extent of human interference, each with several frequent species (Freitag, 1971a; Peinado et al., 1992).

We quantified the extent of variation in leaf properties, and any correlations among them, within four vegetation types with contrasting representation of succulent species: human-induced grasslands dominated by *Stipa tenacissima* on (a) limestone (6 species sampled, none described in the local Flora of Blanca et al. (2011) as having fleshy or succulent leaves), and (b) on deep soils over gypsum (11 species sampled; one leaf succulent), (c) vegetation of shallow soils on gypsum outcrops (5 species sampled; one leaf succulent), and (d) the semi-natural mixture of grass and shrubs on marl (16 species sampled; 4 leaf succulents, 2 shoot succulents).

We addressed nine questions on key aspects of leaf structure and composition in semi-deserts.

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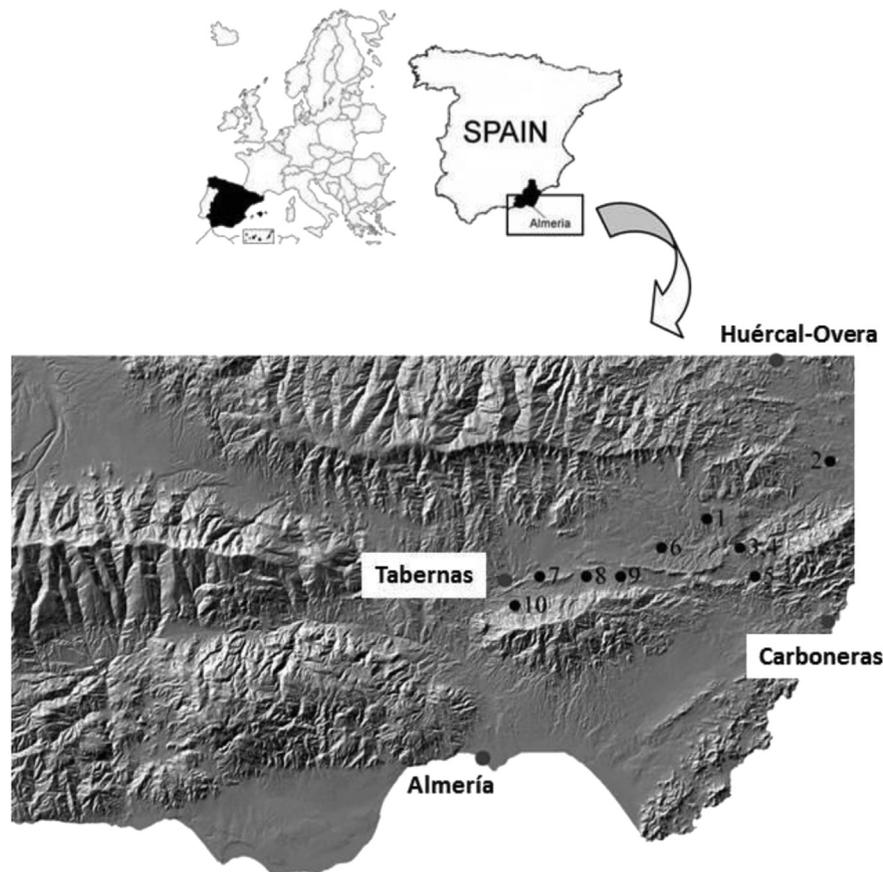


Fig. 1. The study area and the positions of the 10 sites sampled.

1. Is there substantial variation in foliar [N] among species within each of the types of vegetation studied? Foliar [N] is a critically important trait at the world scale (Wright et al., 2004a), and many studies have contrasted formation-types in their mean foliar [N]. For example, in the tropics lowland rain forests have mean foliar [N] 2–3 times greater than those of upper montane rain forests, corresponding to a greater availability of N, and lesser xeromorphy (Grubb, 1977; van de Veg et al., 2009), and in the subtropics low-rainfall woodlands can have mean foliar [N] about twice that of higher-rainfall forests, providing a higher water use efficiency of photosynthesis (Wright and Westoby, 2002). Much less attention has been given to the enormous range of foliar [N] among species of one functional group in a single vegetation type, e.g. up to five-fold variation among deeply shade-tolerant trees of tropical lowland rain forest, often greater than the differences between vegetation types (Grubb, 2002). In the Succulent Karoo semi-desert of South Africa, foliar [N] varied by more than four-fold among 13 shrub species at a single site (8.1–34.6 mg g⁻¹; Carrick, 2001).
2. How far are there similar relationships between specific leaf area and foliar [N] in the various types of vegetation studied? At the world scale specific leaf area (= leaf area/dry mass; SLA) is positively correlated with [N] (Wright et al., 2004a; Osnas et al., 2013). Despite this correlation, SLA and [N] can be independent correlates of light-saturated photosynthetic rate per unit leaf dry mass, A_{\max} , and the correlation of A_{\max} with SLA can be stronger than that with [N] (Reich et al., 1997; Wright et al., 2004b). In a wide-ranging study (rain forest to semi-desert) Reich et al. (1999) compared plants in six climatic regions in terms of the inter-relationships among [N], SLA and A_{\max} ; using logged data they found significant differences in the slopes but not the intercepts.
3. Is SLA correlated with water concentration at saturation (WCS) in the various types of vegetation, and are the concentrations of water and N correlated? In non-succulent plant communities, SLA can be tightly correlated with WCS (Wilson et al., 1999). However, among succulent species, a high WCS reflects degree of succulence (Ogburn and Edwards (2012)), and can coincide with a wide range of SLA (Von Willert et al., 1990; Vendramini et al., 2002). Just as foliar [N] is generally correlated with SLA, it can also be tightly correlated with WCS in vegetation lacking succulent species (Roderick et al., 2000) but this relationship has not been tested for communities including succulents.
4. Do species with longer-lived leaves have lower SLA and/or lower [N] or WCS? There is no doubt that at the world scale species with longer-lived leaves have lower SLA and [N] (Reich et al., 1997, 1999; Wright et al., 2004a; Osnas et al., 2013), but there is little evidence regarding WCS, especially in succulent-rich vegetation.
5. Is SLA correlated with leaf area in any of the types of vegetation? Among species of a given life-form in a particular habitat, leaves can show 'diminishing returns' in photosynthetic area per unit dry mass, i.e., a decline in SLA with increasing leaf size, within species or across species (Marañón and Grubb, 1993; Shipley, 1995; Grubb, 1998; Niklas et al., 2007; Milla and Reich, 2007). However, the opposite trend is often found in comparisons of different vegetation types (Turner, 1994); for example, in the wet tropics mean SLA is lower in the leaves of montane rain forest trees, which are relatively small, than in those of lowland rain forest trees, which are relatively large (Grubb, 1977). There

seems to have been no test of this issue for plants of a given life-form in semi-desert.

6. Is [P] correlated with [N], and what is the range of N/P quotients in each of the vegetation-types? Variation in the foliar N/P quotient has drawn increasing attention as an indicator of the primary limiting nutrient, i.e., for predicting whether plants will respond most strongly to the addition of N or P. Thus, the quotient was found to be an effective indicator of the primary limiting nutrient in freshwater wetland vegetation of the Northern Temperate zone: limited by N if N/P < 14, and by P if N/P > 16 (Koerselman and Meuleman, 1996). Güsewell (2004) reviewed results from a wider range of communities, including the tropical low-rainfall grasslands studied by Penning de Vries et al. (1980), which showed an upper limit of N/P 6 for N-limitation, and a lower limit of N/P 25 for P-limitation. She did not report results from any semi-desert. The relatively few studies of semi-deserts have shown apparently contradictory findings for the usefulness of the N/P quotient to forecast primary nutrient limitation (Table S1 in Supplementary Material). Most N-addition experiments have resulted in increased growth in a number of shrub and perennial herb species and/or ephemeral herbs. The apparent limitation by N in the Mojave Desert was consistent with the mean foliar N/P of 10 found in one study, but not with the quotient of 22 found in another (Table S1). The foliar N/P forecasted primary limitation by N in *Chrysothamnus nauseosus*, and by P in *Sarcobatus vermiculatus*, but an experiment revealed that the reverse was true (Drenovsky and Richards, 2004). We have found no test of limitation by N or P in Eurasian semi-deserts, but the foliar N/P spans the critical interval of 14–16 (Table S1).
7. Are the concentrations of any of the basic cations correlated with WCS, and which basic cations are accumulated in the succulent species? Little is known of the degree that species in a given community accumulate similar basic cations in their vacuoles, or whether this accumulation is approximately in proportion to their WCS values. This question does not seem to have been addressed for most vegetation-types, and has certainly not been addressed for non-saline semi-desert. It is well known that Na is accumulated in many succulents of semi-arid and arid areas (Martínez et al., 2003; Yue et al., 2012), but there has been no previous work on the mineral ions accumulated by succulents in semi-arid Spain.
8. How do the concentrations of various nutrients in whole shoots compare with those found in leaves alone? The attractiveness and value of different plant species to vertebrate herbivores may depend more on the concentrations of nutrients in whole shoots than on those in leaves alone. There has been little work on comparison of nutrient concentrations in leaves and in leaf-bearing stems other than for forest trees. In a study of 20 species in a tropical rain forest, the concentrations of N and Mg

were lower in leaf-bearing twigs, consistent with their lesser development of photosynthetic tissue and greater development of mechanical tissue than in leaves; the mean concentrations of P, K, Na and Ca were similar in leaves and twigs (Grubb and Edwards, 1982).

9. Are there significant and appreciable differences among the four vegetation types in elemental concentrations, and do any such differences parallel differences in soil chemistry? In only a few studies of foliar concentrations in semi-desert plants have comparisons been made among different vegetation-types in one area (e.g. El-Ghonemy et al., 1977) or even populations of a single species in different communities in one area (e.g. Shaltout, 1992). Such differences would be expected based on the strong correlations previously established between soil chemistry and vegetation type in some semi-deserts, e.g. in Australia (Specht, 1972; Beadle, 1981) and South Africa (Grubb, 2005), and the differences among communities in their response to experimental nutrient addition (e.g. Nobel et al., 1988).

We provide a new resolution of the major inter-relationships of leaf structure and function in a highly diverse and understudied ecosystem. Nomenclature follows the Flora Vascular de Andalucía Oriental (Blanca et al., 2011) except that we retain *Stipa* for *S. tenacissima* rather than use *Macrochloa*, and *S. gigantea* rather than use *Celtica*.

2. Materials and methods

2.1. Study area and stands sampled

The study area was bounded by Almería city in the south-west, Tabernas in the north-west, Huércal-Overa in the north-east, and Carboneras in the south-east (Fig. 1). The four stands sampled on limestone were near Sorbas and in Tabernas (Table 1; Fig. 1). The vegetation was dominated by the tall perennial grass *Stipa tenacissima* and contained a scattering of low-stature shrubs such as *Anthyllis cytisoides*, *Rosmarinus officinalis* and *Thymelea hirsuta*. The three stands sampled on marl were in Huércal-Overa and in Tabernas, near Mini-Hollywood. The vegetation was dominated by the perennial grass *Lygeum spartum* and chenopod shrubs such as *Salsola genistoides* and *Salsola oppositifolia*.

Two sites on gypsum were sampled, at Sorbas and near Tabernas (Venta los Yesos). At both sites the soils varied greatly in depth, and we sampled plants in three habitats, making a total of six stands sampled. On shallow soil (0–2 cm) *Sedum gypsicola* was by far the commonest species; on deeper soil (up to c. 10 cm deep) other gypsicolous plants, notably *Coris hispanica*, *Santolina viscosa* and *Teucrium turredanum* were the most abundant; on the deepest soils (>>10 cm) still affected by the gypsum *S. tenacissima* was

Table 1
Details of the study sites where leaves were sampled.

| Site | Location | Latitude | Longitude | Altitude (m asl) | Bedrock | Soil depth (cm) |
|------|-------------------|-----------------|----------------|------------------|------------------------|-----------------------------|
| 1 | Sorbas-Uleila | 37° 09' 33.0" N | 2° 09' 46.3" W | 520 | Limestone | |
| 2 | Huércal-Overa | 37° 22' 56.8" N | 1° 56' 28.5" W | 283 | Marl | |
| 3 | Sorbas | 37° 04' 44.5" N | 2° 05' 45.9" W | 508 | Gypsum | 3.1 0–2; 3.2 0–10; 3.3 >>10 |
| 4 | Sorbas | 37° 04' 48.1" N | 2° 05' 44.7" W | 441 | Limestone/conglomerate | |
| 5 | Sorbas-Carboneras | 37° 04' 24.2" N | 2° 03' 41.2" W | 454 | Limestone | |
| 6 | Alhamilla | 37° 01' 57.6" N | 2° 23' 43.0" W | 430 | Limestone | |
| 7 | Tabernas | 37° 02' 16.4" N | 2° 25' 08.4" W | 326 | Marl | |
| 8 | Tabernas | 37° 02' 15.4" N | 2° 25' 11.9" W | 409 | Marl | |
| 9 | Venta los Yesos | 37° 04' 59.7" N | 2° 17' 43.3" W | 566 | Gypsum | 9.1 0–2; 9.2 0–10; 9.3 >>10 |
| 10 | Tabernas | 37° 01' 07.0" N | 2° 25' 53.0" W | 350 | Marl | |

dominant, and shrubs such as *Rosmarinus officinalis* and *Hammada articulata* were found as well as some gypsicolous species. In presenting our results we treat together data for species in the first two habitats.

2.2. Soils

In each stand a bulked sample of topsoil (0–20 cm) was taken at three random points and mixed as one combined sample per stand. Samples were transported to the lab and air-dried, crushed and sieved (<2 mm). The pH was determined in a 1:2.5 soil: CaCl₂ 0.01 M solution. Total organic nitrogen was measured by the Kjeldahl method; available P by the Olsen method; available potassium, calcium and magnesium by extraction with ammonium acetate; electrical conductivity (EC) and soluble ions (Na and SO₄) by measurements of soil saturation extracts (after García et al. (2002)).

The study sites varied strongly in nutrient availability (Table 2). Thus, soils on marl had appreciably lower total nitrogen concentrations than both shallow and deep soils over gypsum, and lower N and P than those on limestone. Soils on marl and limestone had similar concentrations of exchangeable K, Mg and Ca. As expected, the soils of various depths over gypsum had appreciably higher concentrations of exchangeable Ca.

Soils on marl and limestone overlapped in pH, but soils on gypsum had slightly lower values (Table 2). Soil electrical conductivity increased along the sequence limestone, marl, gypsum, in part reflecting differences in SO₄²⁻ concentration, which was much lower on limestone than on marl or gypsum, with no significant difference between the ranges for marl and gypsum (Table 2). By contrast, shallow soils on gypsum outcrops had the lowest values for Na⁺ concentration in soil solution, with soils on limestone intermediate and those on marl highest.

The multivariate structure of the 9 soil variables at the 10 sites (14 samples including gypsum microhabitats) was studied by principal component analysis (PCA). The first PCA axis (explaining 51.4% of variance) separated 'Limestone' sites from 'Gypsum' sites, with highest pH and Mg values at 'Limestone' sites and highest electrical conductivity (EC), SO₄ and Ca at 'Gypsum' sites (Appendix

Fig. S1 in Supplementary Material). The second PCA axis (17.5% of variance) distinguished the 'Marl' sites (intermediate on the first gradient) because of their high values of Na in the soil solution.

2.3. Leaf properties

At site 1 we sampled only the dominant *Stipa tenacissima*, and at sites 2–9 we sampled the common perennial species. At site 8 we additionally sampled the leaf-succulent annual *Mesembryanthemum nodiflorum*, typical of certain communities on marl (Peinado et al., 1992).

In April 2000 between 10:00 and 18:00 h we collected healthy shoots from three individuals of each species at each site in each vegetation type. These were transported to the lab in self-seal plastic bags with moist paper, and rehydrated overnight with shoot bases under water, covered with plastic lined with wet filter paper. For each individual, we determined the mass of 10–20 mature healthy leaves with an analytical balance (± 1 mg) immediately after excision from the hydrated shoots.

We used leaves from the most recent flush that was mature, and the age of this flush varied among species. For 13 dicot species we sampled mature leaves apparently developed in the current year (Table 3; groups 1, 2 and 3). For 10 species (group 4) the leaves in the younger flush were not mature and we sampled leaves apparently developed in the previous year and without signs of senescence. When relating trait values to leaf longevity, we assumed that the species in groups 1 and 2 had shorter-lived leaves than those in group 4.

For three species with minute leaves (group 5) we used current whole shoots. For the two grasses we used only the blades, and excluded the terminal 5 cm which was already senescing on otherwise healthy leaves apparently of the current year. Leaves of *Cistus clusii* were sampled evenly along the full length of the leaf-bearing shoots.

Sampled leaves from each individual were scanned and their areas determined with image analysis software (MideBMP, Almería) before oven drying at 70 °C for 48 h, after which dry mass was determined. Leaves of most dicot species had almost no petiole;

Table 2
Selected chemical properties of the topsoil at the sites where leaves were sampled; bold type indicates outstanding values.

| | N | P | K | Ca | Mg | pH | EC | Na | SO ₄ |
|----------------------------|--------------------|--------------------|--------------------|--------------------|--------------------|----------------------|--------------------|---------------------|---------------------|
| | mg g ⁻¹ | μg g ⁻¹ | μg g ⁻¹ | mg g ⁻¹ | μg g ⁻¹ | (CaCl ₂) | dS m ⁻¹ | meq l ⁻¹ | meq l ⁻¹ |
| Sites on marl | | | | | | | | | |
| 2. Huércal | 0.08 | 1.6 | 120 | 8.1 | 208 | 7.8 | 4.1 | 5.76 | 25.9 |
| 7. Tabernas | 0.08 | 3.9 | 171 | 4.9 | 133 | 8.0 | 4.9 | 1.31 | 35.5 |
| 8. Tabernas | 0.05 | 4.7 | 51 | 7.6 | 130 | 7.8 | 7.0 | 4.59 | 56.7 |
| 10. Tabernas | 0.06 | 2.6 | 85 | 5.3 | 178 | 8.1 | 2.6 | 3.42 | 13.9 |
| Sites on gypsum | | | | | | | | | |
| (a) Very shallow outcrops | | | | | | | | | |
| 3a. Sorbas | 0.21 | 2.3 | 87 | 16.6 | 108 | 7.5 | 6.0 | 0.70 | 24.1 |
| 9a. Tabernas | 0.24 | 8.1 | 139 | 17.3 | 83 | 7.2 | 6.4 | 0.65 | 45.0 |
| (b) Average depth outcrops | | | | | | | | | |
| 3b. Sorbas | 0.06 | 0.0 | 15 | 16.7 | 50 | 7.6 | 6.4 | 0.80 | 44.1 |
| 9b. Tabernas | 0.08 | 0.0 | 105 | 15.9 | 53 | 7.4 | 6.8 | 0.40 | 61.4 |
| (c) Deeper soils | | | | | | | | | |
| 3c. Sorbas | 0.31 | 0.8 | 75 | 16.6 | 155 | 7.6 | 3.4 | 2.76 | 18.8 |
| 9c. Tabernas | 0.31 | 6.2 | 22 | 12.4 | 63 | 7.6 | 6.7 | 0.73 | 60.2 |
| Sites on limestone | | | | | | | | | |
| 1. Sorbas-Uleila | 2.30 | 6.5 | 120 | 7.4 | 148 | 8.2 | 0.7 | 0.90 | 0.9 |
| 4. Sorbas | 1.80 | 5.7 | 127 | 7.8 | 280 | 7.8 | 3.5 | 2.63 | 3.8 |
| 5. Sorbas-Carboneras | 1.30 | 5.5 | 145 | 6.9 | 180 | 8.2 | 1.3 | 1.36 | 3.3 |
| 6. Tabernas -Alhama | 0.15 | 4.4 | 143 | 5.4 | 158 | 8.3 | 1.3 | 1.25 | 3.3 |

Artemisia barrelieri and *S. viscosa* had petioles of appreciable length relative to that of the whole leaf, and these were included in our samples. The dried leaves were ground to a fine powder.

Concentrations of N, P, K, Na, Ca and Mg were determined by standard methods described by Grubb et al. (1994). With respect to cation concentrations, given that there is no standard definition, we considered as 'accumulator species' those in which the concentration of a given ion is more than twice the maximum value of the species in the grassland on limestone.

2.4. Data analysis

Relationships between leaf traits were tested by correlation analysis. We used Student's t-test to compare groups based on leaf longevity, vegetation type and whether or not Blanca et al. (2011) reported the species to be 'nitrophilous', i.e. markedly responsive to soil-enrichment by animals. All variables were tested for normality and heteroscedasticity before analysis and transformed if necessary (for standard statistical tests see Sokal and Rohlf, 1987). For analyses we used Statistica v. 8.0 software (Stat Soft, Tulsa, OK, USA).

3. Results

3.1. Range of N concentration in leaves

Foliar [N] varied by 5-fold in three of the four vegetation types sampled, i.e., among the 6 species of grassland on limestone (from a mean value of 4.3 mg g⁻¹ for *Stipa tenacissima* to 21.8 mg g⁻¹ for *Anthyllis cytisoides*), among the 11 species of grassland on deep soil over gypsum (from 4.8 mg g⁻¹ for *Sedum sediforme* to 23.1 mg g⁻¹ for *Lepidium subulatum*), and among the 16 species of vegetation on marl (from 6.6 mg g⁻¹ for *Arthrocnemum macrostachyum* to 33.5 mg g⁻¹ for *Sideritis pusilla*; Table 4). Excluding grasses, highly succulent species, and species with shoots bearing scale leaves, foliar [N] varied by 2.5-, 3.2- and 3.5-fold in the three vegetation types respectively. The total range for the five species sampled on shallow soils over gypsum was only 2.4-fold (from 8.1 mg g⁻¹ for *S. gypsicola* to 19.6 mg g⁻¹ for *S. viscosa*). There was an 8-fold range across the semi-desert species pooling across all vegetation types (from 4.3 mg g⁻¹ in *Stipa tenacissima* in limestone grassland to 33.5 mg g⁻¹ in *Sideritis pusilla* on marl).

3.2. SLA related to the concentration of N

SLA was positively correlated with foliar [N] in each of the four vegetation types sampled (Fig. 2). The two variables were very tightly correlated in the grassland on limestone ($r = 0.99$) and tightly correlated in the grassland on deep soil over gypsum ($r = 0.89$). The succulent *S. sediforme* did not have a high SLA relative to its foliar [N], and the correlation in the grassland on deep soil over gypsum was weakened slightly if *S. sediforme* was excluded ($r = 0.88$). On shallow soil over gypsum, *S. gypsicola* had a high SLA relative to its foliar [N], but the four remaining species had a tight correlation between the two variables ($r = 0.95$). In the vegetation on marl there was a much weaker correlation between SLA and [N] ($r = 0.66$; $P = 0.013$).

3.3. SLA related to the concentration of water

The relationship between SLA and WCS varied among vegetation types (Fig. 3). In the grassland on limestone there was a strong correlation (Fig. 3a; $r = 0.96$); also WCS was significantly correlated with [N] (Fig. 4a; $r = 0.97$). The leaf succulent *S. sediforme* had a high WCS relative to its SLA (Fig. 3b); when that species was excluded,

Table 3

The flushes of leaves found on the dicotyledonous species studied, and the flushes sampled for analysis. The authorities for grass names are as follows: *Stipa tenacissima* L. and *Lygeum spartum* L.

- Species on which there were apparently mature leaves in the current flush (presumed to have grown in the current year, sampled for analysis), and the primary leaves of the previous year had all or mostly fallen (axillary shoots were growing out on the older stems in those species marked by an asterisk)**

Anthyllis cytisoides L.*
Anthyllis terniflora (Lag.) Pau*
Artemisia barrelieri Besser*
Frankenia thymifolia Desf.*
Helianthemum alypoides Losa & Rivas Goday*
Lepidium subulatum L.*
Salsola genistoides Poirlet
S. oppositifolia Desf.
Sideritis pusilla (Lange) Pau*
Suaeda cf. vera J. F. Gmelin

- Species on which there were apparently mature leaves in the current flush (presumed to have grown in the current year), while the leaves of the previous year were apparently dead but still attached**

Gypsophila struthium L.
Santolina viscosa Lag.

- Species on which the leaves in the younger flush appeared to be mature (sampled, presumed to have grown in the current year), and there were many apparently living leaves in an older flush (not sampled, presumed to be from the previous year)**

Helianthemum almeriense Pau

- Species on which the leaves of the younger flush had not reached full length (not sampled) and there were many apparently fully healthy mature leaves in the older flush (sampled and presumed to be of the previous year)**

(a) *New leaves mostly more than half the length of mature leaves*
Coris hispanica Lange (some leaves of 1998 flush present)

Phlomis purpurea L.
Rosmarinus eriocalyx Jord. & Fourr.
R. officinalis L.

Sedum sediforme (Jacq.) Pau
Thymelaea hirsuta (L.) Endl.
Teucrium turredanum Losa & Rivas Goday

(b) *New leaves all or mostly less than half full length*
Helianthemum squamatum (L.) Dum. Cours.
Lycium intricatum Boiss. (a few leaves of 1998 flush present)
Sedum gypsicola Boiss. & Reut.

- Leaves scale-like; whole shoots of the current year were processed**

Anabasis articulata (Forssk.) Moq.
Arthrocnemum macrostachyum (Moric.) Moris
Hammada articulata (Moq.) O. Bolos & Vigo

- Apparently only one flush of leaves but not convincingly that of the current year; hard to interpret**

Cistus clusii Dunal

the relationship between SLA and WCS in the grassland on deep soil over gypsum was similar to that for grassland on limestone but with more scatter (Fig. 3b; $r = 0.72$; $P = 0.03$). However, the correlation between foliar [N] and WCS was weak even when *S. sediforme* was excluded ($r = 0.50$); by the standards of the majority of species *Hammada articulata* and *Lepidium subulatum* had low values for WCS at high values of N concentration (Table 4). The remaining eight species showed a tight correlation like that for the grassland species on limestone (Fig. 4b; $r = 0.95$). Among the species on shallow soil over gypsum, excluding the leaf succulent *S. gypsicola*, there was a tight correlation of SLA with WCS (Fig. 3c; $r = 0.97$) and between WCS and [N] (Fig. 4c; $r = 0.94$).

In the vegetation on marl there was only a non-significant empirical trend for higher SLA with higher WCS (Fig. 3d; $r = 0.23$). The relationship between WCS and [N] was different for the species described as leaf-succulent by Blanca et al. (2011) and for the rest – the graminoids, stem-succulents, 'somewhat succulent' species and non-succulents (Fig. 4d). For the four leaf succulents WCS showed a non-significant trend to decline with increasing [N] ($r = 0.82$; $P = 0.17$), while for the remaining 12

Table 4

The mean values for lamina area, specific leaf area, water concentration at saturation and concentrations of nutrients in leaves of species from various communities; the species are ordered within each community according to increasing N concentration; concentrations are rounded off to two or three significant figures; values in bold type indicate that the species is an 'accumulator' of the element in question according to the test given in the text; the letters indicating succulence (in the column Succ) refer to the descriptions in the *Flora of Blanca et al. (2011)*: LS leaves succulent or fleshy, LSS leaves somewhat succulent, SS shoot succulent, SSS shoot somewhat succulent. Species which invade old-fields first are marked with asterisks after their name.

| | | Succ | Mean area of leaf | Mean SLA | Mean water per unit dry mass | Mean N concn | Mean P concn | Mean K concn | Mean Na concn | Mean Ca concn | Mean Mg concn | N/P | Sites at which sampled |
|---------------------------------------------|-----------------|------|----------------------|------------------------------------|---------------------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|------|------------------------------|
| | | | (mm ²) | (cm ² g ⁻¹) | (g g ⁻¹) | (mg g ⁻¹) | (mg g ⁻¹) | (mg g ⁻¹) | (mg g ⁻¹) | (mg g ⁻¹) | (mg g ⁻¹) | | |
| Grassland on limestone | | | | | | | | | | | | | |
| <i>Stipa tenacissima</i> | Poaceae | | ND | 29 | 1.70 | 4.31 | 0.47 | 6.2 | 0.21 | 1.5 | 0.4 | 9.2 | 1, 4, 5, 6 |
| <i>Thymelaea hirsuta*</i> | Thymelaeaceae | LSS | 5.8 | 46 | 2.60 | 8.67 | 0.9 | 11 | 1.1 | 15 | 3 | 8.6 | 5 & 6 |
| <i>Rosmarinus officinalis</i> | Lamiaceae | | 31 | 48 | 2.40 | 9.15 | 0.49 | 17 | 0.91 | 21 | 2.3 | 18.7 | 4 |
| <i>Cistus clusii</i> | Cistaceae | | 35 | 57 | 2.70 | 11.5 | 0.68 | 7.3 | 0.59 | 21 | 2.9 | 16.9 | 4 & 6 |
| <i>Phlomis purpurea</i> | Lamiaceae | | 61 | 89 | 3.20 | 18.8 | 1.33 | 15 | 1.6 | 27 | 2.2 | 14.1 | 5 |
| <i>Anthyllis cytisoides*</i> | Fabaceae | | 39 | 101 | 3.80 | 21.8 | 1.21 | 25 | 0.28 | 29 | 6.5 | 18 | 6 |
| Grassland on deep soil over gypsum | | | | | | | | | | | | | |
| <i>Sedum sediforme</i> | Crassulaceae | LS | 65 | 37 | 9.30 | 4.76 | 0.63 | 4.9 | 0.22 | 57 | 1.5 | 7.6 | 3, 3 |
| <i>Stipa tenacissima</i> | Poaceae | | ND | 25 | 1.60 | 5.07 | 0.24 | 4.9 | 0.3 | 2 | 0.46 | 21.1 | 3, 3 & 9.3 |
| <i>Rosmarinus eriocalyx</i> | Lamiaceae | | 23 | 48 | 2.10 | 7.26 | 0.48 | 9.3 | 0.91 | 15 | 3 | 15.1 | 9.3 |
| <i>Thymelaea hirsuta*</i> | Thymelaeaceae | LSS | 2.9 | 56 | 2.60 | 10.2 | 0.44 | 5.8 | 1.1 | 19 | 1.5 | 23.2 | 3, 3 |
| <i>Rosmarinus officinalis</i> | Lamiaceae | | 32 | 39 | 2.60 | 10.6 | 0.8 | 11 | 1.8 | 17 | 1.9 | 13.3 | 3, 3 |
| <i>Helianthemum alypoides</i> | Cistaceae | LSS | 25 | 72 | 3.60 | 14 | 0.86 | 5.7 | 0.42 | 16 | 3.5 | 16.3 | 3, 3 & 9.3 |
| <i>Gypsophilla struthium</i> | Caryophyllaceae | | 29 | 122 | 4.30 | 16 | 0.63 | 12 | 0.7 | 74 | 8.2 | 25.4 | 3, 3 & 9.3 |
| <i>Coris hispanica</i> | Myrsinaceae | | 17 | 69 | 4.60 | 16.5 | 0.46 | 7.1 | 0.97 | 50 | 2.1 | 35.9 | 3, 3 |
| <i>Anthyllis terniflora*</i> | Fabaceae | | 45 | 137 | 4.70 | 22.6 | 0.84 | 11 | 1.8 | 44 | 3.2 | 26.9 | 3, 3 & 9.3 |
| <i>Hammada articulata*</i> | Chenopodiaceae | SSS | ND | ND | 2.50 | 23 | 0.97 | 28 | 0.87 | 19 | 5.2 | 23.7 | 9.3 |
| <i>Lepidium subulatum</i> | Brassicaceae | | 24 | 108 | 2.50 | 23.1 | 0.8 | 15 | 0.84 | 24 | 3.6 | 28.9 | 9.3 |
| Open vegetation on shallow soil over gypsum | | | | | | | | | | | | | |
| <i>Sedum gypsicola</i> | Crassulaceae | LS | 24 | 86 | 13.30 | 8.14 | 1.14 | 12 | 0.3 | 44 | 2.8 | 7.1 | 3, 1 & 9.1 |
| <i>Teucrium turredanum</i> | Lamiaceae | | 23 | 47 | 2.60 | 10.3 | 0.45 | 16 | 0.69 | 18 | 2.6 | 22.9 | 3, 2 |
| <i>Coris hispanica</i> | Myrsinaceae | | 14 | 81 | 4.40 | 16.1 | 0.34 | 12 | 1.2 | 45 | 2 | 47.4 | 3, 2 & 9.2 |
| <i>Helianthemum squamatum</i> | Cistaceae | | 84 | 68 | 4.00 | 16.2 | 0.5 | 7.7 | 11 | 35 | 5.5 | 32.4 | 3, 2 & 9.2 |
| <i>Santolina viscosa</i> | Asteraceae | | 29 | 90 | 4.50 | 19.6 | 0.99 | 20 | 7.8 | 33 | 3.8 | 19.8 | 3, 2 & 9.2 |
| Vegetation on marl | | | | | | | | | | | | | |
| <i>Arthrocnemum macrostachyum</i> | Chenopodiaceae | | ND | ND | 2.70 | 6.63 | 0.54 | 19 | 29 | 42 | 11 | 12.3 | 8 |
| <i>Stipa tenacissima</i> | Poaceae | | ND | 39 | 1.70 | 7.96 | ND | 9.1 | 0.16 | 1.4 | 0.71 | ND | 2 |
| <i>Anabasis articulata</i> | Chenopodiaceae | SS | ND | ND | 2.70 | 8.95 | 0.48 | 23 | 20 | 60 | 14 | 18.6 | 10 |
| <i>Frankenia thymifolia</i> | Frankeniaceae | | 2.9 | 39 | 1.70 | 9.64 | 0.51 | 4.5 | 17 | 54 | 9.3 | 18.9 | 8 |
| <i>Thymelaea hirsuta*</i> | Thymelaeaceae | LSS | 6 | 69 | 2.40 | 9.92 | 0.75 | 8.4 | 2.1 | 12 | 4 | 13.2 | 2 & 8 |
| <i>Lygeum spartum</i> | Poaceae | | ND | 33 | 2.60 | 10.2 | 0.58 | 34 | 1.2 | 5.2 | 1.9 | 17.6 | 2, 7, 8 & 10 |
| <i>Lycium intricatum</i> | Solanaceae | LS | 25 | 79 | 7.00 | 10.4 | 0.77 | 55 | 62 | 31 | 19 | 13.5 | 8 |
| <i>Suaeda cf. vera</i> | Chenopodiaceae | LS | 7.1 | 54 | 6.10 | 14.2 | 0.71 | 19 | 140 | 3.6 | 2.7 | 20 | 8 |
| <i>Euzomodendron bourgaeum</i> | Brassicaceae | LS | 68 | 91 | 6.60 | 15.3 | 1.1 | 30 | 18 | 98 | 17 | 13.9 | 10 |
| <i>Hammada articulata*</i> | Chenopodiaceae | SSS | ND | ND | 2.60 | 23 | 1.6 | 36 | 0.84 | 27 | 8.3 | 14.4 | 7 & 8 |
| <i>Salsola oppositifolia</i> | Chenopodiaceae | LS | 24 | 51 | 5.90 | 23.5 | 0.93 | 64 | 54 | 28 | 9.5 | 25.3 | 2 & 8 |
| <i>Artemisia barrelieri*</i> | Asteraceae | | 19 | 111 | 3.30 | 24.1 | 1.12 | 29 | 5.2 | 18 | 3 | 21.5 | 2 & 7 |
| <i>Helianthemum almeriense</i> | Cistaceae | LSS | 12 | 110 | 4.00 | 25.9 | 0.62 | 14 | 0.41 | 11 | 3.8 | 41.8 | 2 |
| <i>Salsola genistoides</i> | Chenopodiaceae | | 5.7 | 74 | 4.10 | 28.5 | 0.48 | 36 | 28 | 18 | 13 | 59.4 | 2, 8, 10 |
| <i>Anthyllis terniflora*</i> | Fabaceae | | 40 | 101 | 3.90 | 28.6 | 1.03 | 19 | 5.7 | 35 | 8.3 | 27.8 | 8 |
| <i>Sideritis pusilla</i> | Lamiaceae | | 13 | 93 | 2.60 | 33.5 | 1.02 | 40 | 1.2 | 25 | 4.2 | 32.8 | 2 |

species there was a significant positive correlation ($r = 0.68$; $P = 0.015$).

3.4. SLA and concentrations of N and of water in leaves of differing longevity

The only vegetation type in which a balanced comparison can be made between species with short- and long-lived leaves (cf. Tables 3 and 4) is the grassland on deeper soils over gypsum, where we sampled four species with short-lived leaves and five with long-lived. The short-lived leaves had a markedly higher SLA (mean \pm standard error of 110 ± 13.9 vs 50 ± 4.4 cm² mg⁻¹; $t = 4.31$; $P = 0.004$) and a markedly higher [N] (18.9 ± 2.31 vs 9.9 ± 1.96 mg g⁻¹, $t = 3.01$; $P = 0.02$) but there was no significant difference in WCS (3.8 vs 4.2 g g⁻¹; 3.8 vs 3.0 g g⁻¹ excluding the

leaf succulent *S. sediforme*). In the grassland on limestone the one species with short-lived leaves (*Anthyllis cytisoides*) had the highest SLA, [N] and WCS. In the vegetation on shallow soils over gypsum the same was found for SLA and [N] but not WCS. In the vegetation on marl there were 8 dicots with short-lived leaves and only 2 with long-lived; the former had a much higher [N] (23.5 ± 2.79 vs 10.2 ± 0.25 mg g⁻¹; $t = 2.28$; $P = 0.05$) but a statistically similar SLA (75 vs 74 cm² g⁻¹) and WCS (3.9 vs 4.7 g g⁻¹).

When the dicots in all four vegetation types were lumped, averaging for given species across vegetation types, the difference in [N] between species with short-lived and long-lived leaves was appreciable and significant (21.1 ± 1.96 vs 11.2 ± 1.29 mg g⁻¹; $t = 3.97$; $P = <0.001$) and also that in SLA (86.2 ± 8.06 vs 63.0 ± 5.43 cm² g⁻¹; $t = 2.23$; $P = 0.037$). There was no significant difference in mean WCS, even when the two leaf succulent *Sedum*

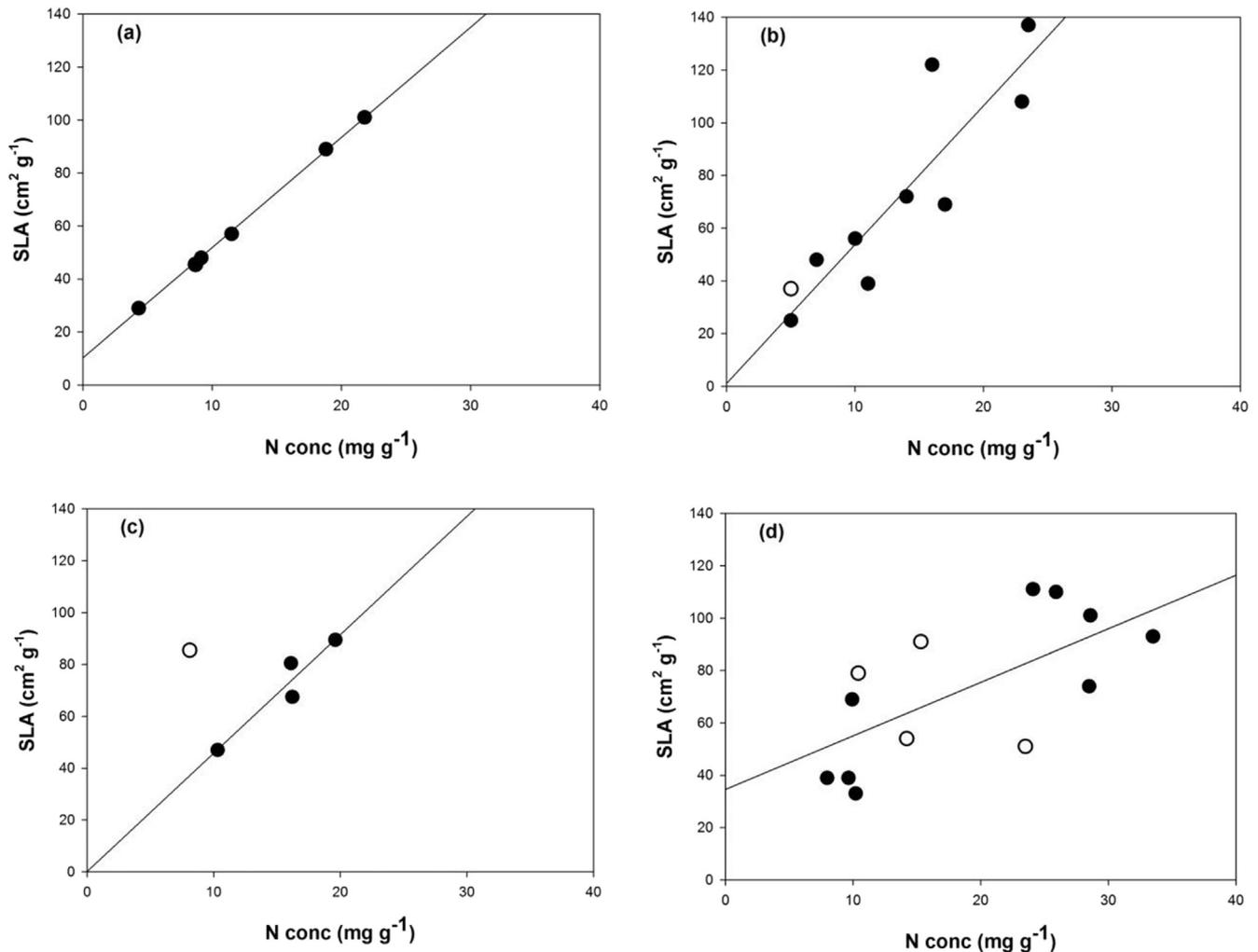


Fig. 2. Relationships of specific leaf area to foliar nitrogen concentration in different kinds of semi-desert: (a) grassland on limestone, (b) grassland on deep soil over gypsum, (c) vegetation on shallow soil over gypsum, and (d) vegetation on marl. The regression line in (c) omits one outlier (*Sedum gypsicola*). Empty circles indicate leaf-succulents, and filled circles other species.

species were omitted from the analysis (3.9 ± 0.37 vs 3.6 ± 0.22 g g^{-1} ; $t = 0.52$; $P = 0.61$).

3.5. SLA and concentrations of N and of water in leaves of differing area

Leaf area was statistically independent of both SLA and foliar [N] in each of the vegetation types studied. There was only one vegetation type in which there was a significant correlation between leaf area and WCS: in grassland on deep soils over gypsum leaves of larger area had higher WCS ($r = 0.80$; $P = 0.01$).

3.6. Correlations between [N] and [P] and N/P quotients

The foliar nutrient concentrations of species in all four vegetation types are shown in Table 4, and their correlations in Table 5. Foliar [N] and [P] were positively correlated for the species in grassland on limestone and for the species in grassland on deeper soils over gypsum (Fig. 5a and b), but not for plants on shallow soils over gypsum or for vegetation on marl (Fig. 5c and d). For the vegetation on marl, there was an apparent wedge-shaped relationship with a wide range of [P] at high [N] but not at low N (Fig. 5d).

The N/P quotient varied across species within vegetation types, and the ranges varied across the vegetation types from a mean value of 9.2 for *Stipa tenacissima* to 18.7 for *Rosmarinus officinalis* in grassland on limestone with a mean across species of 14.3; from 7.6 for *S. sediforme* to 35.9 for *C. hispanica* in grassland on deep soil over gypsum, with a mean across species of 21.6; from 7.1 for *S. gypsicola* to 47.4 for *C. hispanica* in open vegetation on shallow soils over gypsum with a mean across species of 25.9; and from 12.3 for *Arthrocnemum macrostachyum* to 59.4 for *Salsola genistoides* in vegetation on marl with a mean across species of 23.4; only 5 of 15 species had a quotient <16.

3.7. WCS and concentrations of basic cations

For the species in grassland on limestone, WCS was positively correlated with [Ca] and [Mg], and increased empirically with [K] ($P = 0.072$), and was statistically independent of [Na] (Table 5). For the species in grassland on deeper soils over gypsum WCS was positively correlated with [Ca], but not with [K], [Mg] or [Na]. For plants on marl WCS was correlated with [Na] but not [K], [Ca] or [Mg]. For plants on shallow soils over gypsum, WCS was not correlated with the concentration of any major basic cation.

Among species sampled in marl vegetation Na was the ion

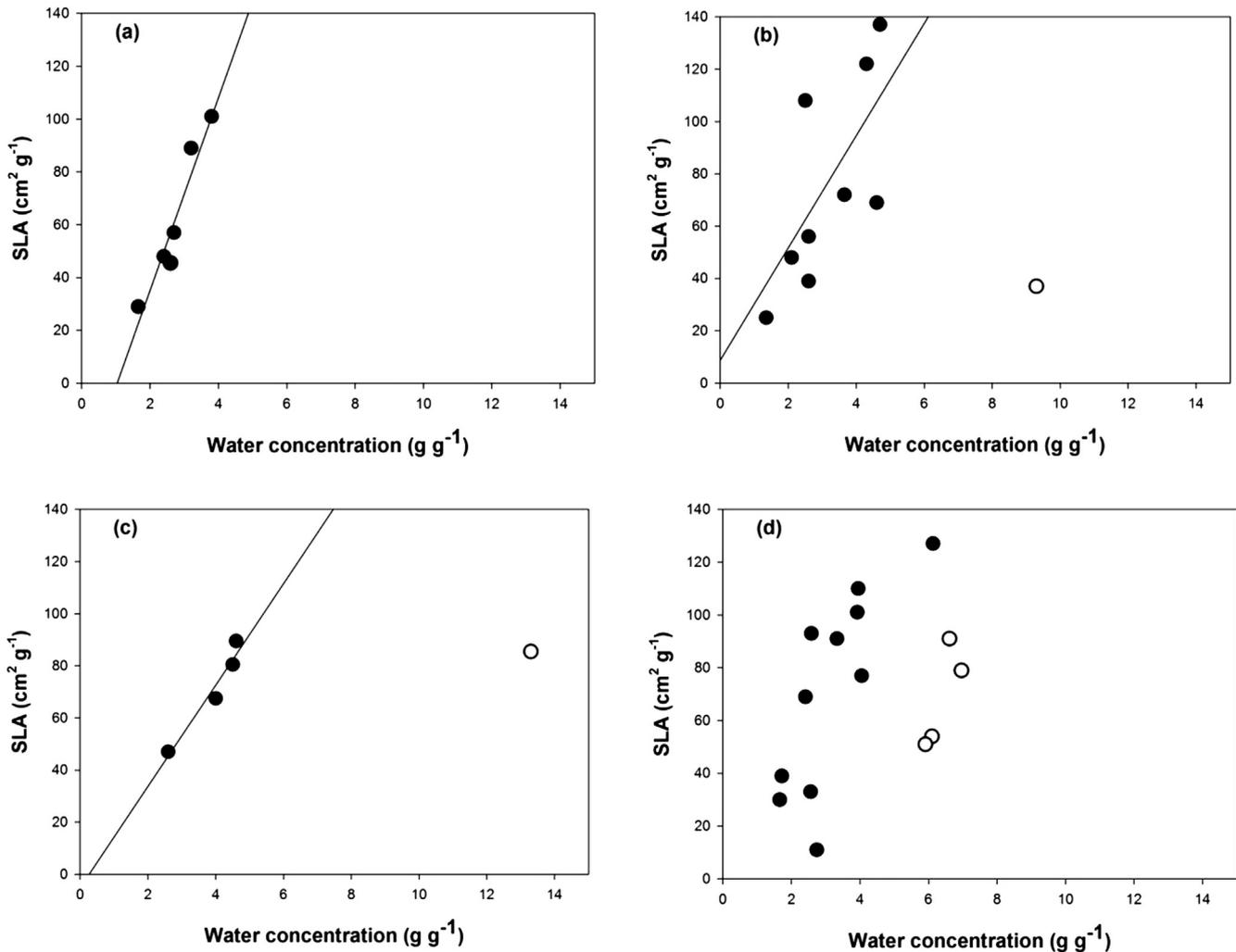


Fig. 3. Relationships of specific leaf area to foliar water concentration at saturation in different kinds of semi-desert: (a) grassland on limestone, (b) grassland on deep soil over gypsum, (c) vegetation on shallow soil over gypsum, and (d) vegetation on marl. The regression lines in (b) and (c) omit one outlier (*Sedum sediforme* and *S. gypsicola* respectively). Empty circles indicate leaf-succulents, and filled circles other species.

'accumulated' (see Methods) by the greatest number of species (10/16). One perennial species accumulated Na to $>100 \text{ mg g}^{-1}$ (*Suaeda cf. vera*), and the annual *M. nodiflorum* accumulated an even higher concentration of Na (180 mg g^{-1}). Four species accumulated Mg (*Anabasis articulata*, *Euzomodendron bourgaeum*, *Lycium intricatum* and *Salsola genistoides*), while two accumulated Ca (*A. articulata* and *Euzomodendron bourgaeum*), and two accumulated K (*Lycium intricatum* and *Salsola oppositifolia*).

Among the five species on shallow soil over gypsum there were only two Na accumulators (*Helianthemum squamatum* and *S. viscosa*), and there were no K, Ca or Mg accumulators. In the grassland on deeper soil over gypsum there was one Ca accumulator (*Gypsophila struthium*) and no K, Na or Mg accumulators.

3.8. Comparison of nutrient concentrations in whole shoots and leaves

The nutrient concentrations of whole shoots and leaves alone are compared in [Appendix Table S2](#). For N and P the concentrations in whole shoots and leaves alone were similar. For N there was a strong correlation of leaf and shoot concentrations across species when the two samples of *Salsola genistoides* were omitted

($r = 0.97$), and for P when the values for a single collection of *Salsola oppositifolia* were omitted ($r = 0.81$).

For K the whole-shoot concentrations were significantly lower than in the leaves alone (significant t values for *Stipa tenacissima*, *Salsola* spp. and across species), and there was a strong correlation across species ($r = 0.90$ for a straight-line plot and $r = 0.94$ for exponential). For Na, the whole-shoot concentrations were generally similar to those in the leaves alone, and the values were correlated across species for the whole data set ($r = 0.96$) but most values were clustered in the lower-concentration range ($<2 \text{ mg g}^{-1}$) and for these the correlation was also relatively strong ($r = 0.75$).

For Mg the whole-shoot concentrations were slightly higher than those in the leaves alone (significant only for *Lygeum spartum*); there was a strong correlation across species over the whole range of concentrations ($r = 0.84$), and an even stronger one for the 12 samples with $<3.0 \text{ mg g}^{-1}$ in the leaves ($r = 0.96$). For Ca, values were lower in the shoots than in leaves alone (significant for *Lygeum spartum* and *Rosmarinus officinalis*) but the reverse trend for *Stipa tenacissima*; there was a strong correlation when the whole collection of values was considered ($r = 0.93$), but none for the 10 samples with $<4.0 \text{ mg g}^{-1}$ in the leaves ($r = -0.33$, trend negative).

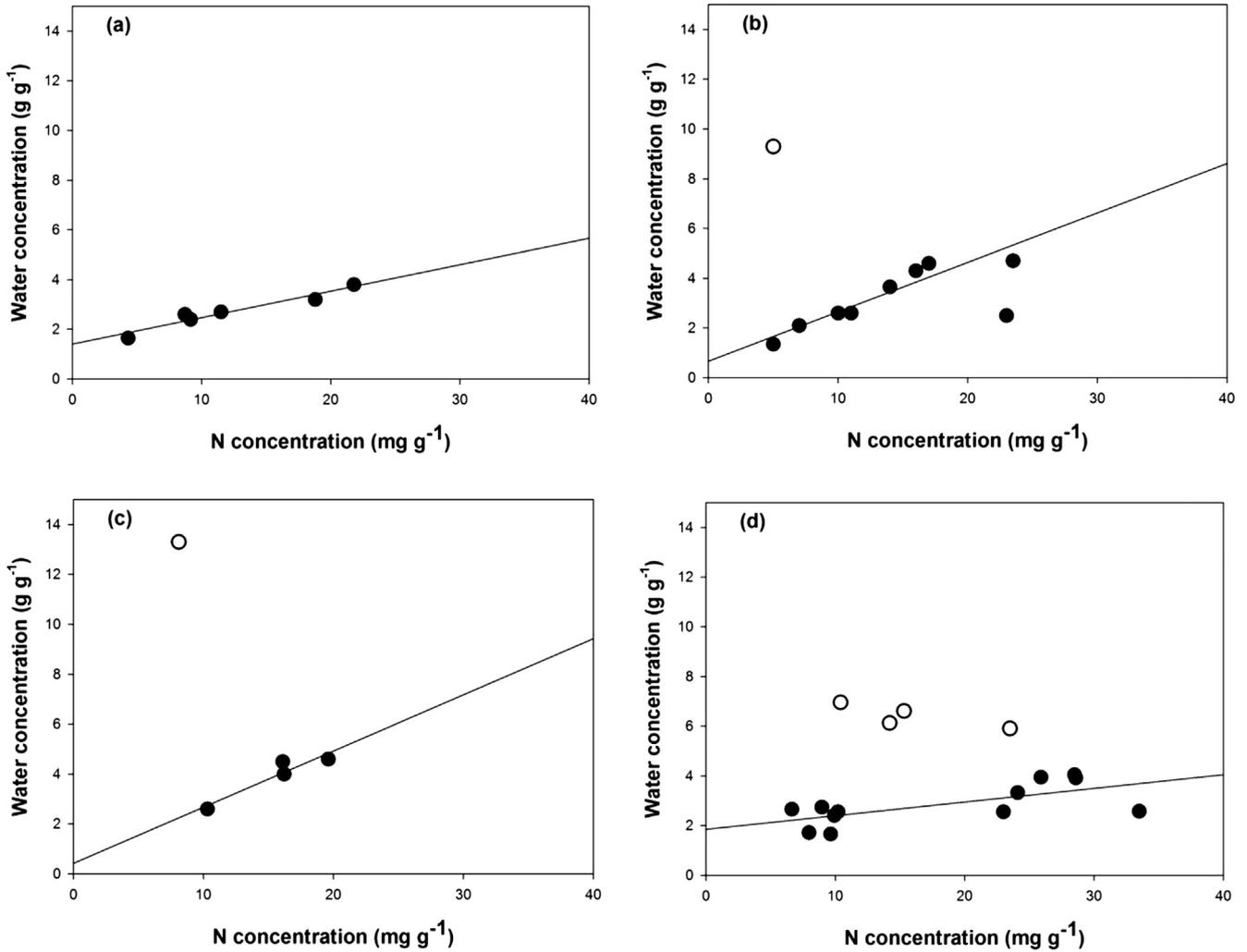


Fig. 4. Relationships of foliar water concentration at saturation to foliar nitrogen concentration in different kinds of semi-desert: (a) grassland on limestone, (b) grassland on deep soil over gypsum, (c) vegetation on shallow soil over gypsum, and (d) vegetation on marl. The regression line in (b) omits three outliers (*Sedum sediforme*, *Hammada articulata* and *Lepidium subulatum*, the last two species coincident on one point), that in (c) omits one outlier (*Sedum gypsicola*). Empty circles indicate leaf-succulents, and filled circles other species.

3.9. Differences among vegetation types in mean foliar concentrations, and possible relationship to differences in soil chemistry

None of the differences among vegetation types in foliar [N] or [P] is significant (Appendix Table S3). The vegetation on shallow

soil over gypsum has a significantly higher mean [Ca] than the grassland on limestone; the mean [Ca] values of the grassland on deeper soil over gypsum and the vegetation on marl are close to that for the vegetation on shallow soil over gypsum (31 and 29 vs 35 mg g⁻¹), but the variance is so great that the difference from the [Ca] of the grassland on limestone (19 mg g⁻¹) is not significant.

Table 5

Correlations (r values) between concentrations of nutrients and of water in leaves of species in the various communities; values in bold type if *P* < 0.05, and in bold italic type if *P* < 0.01.

| | Total | Grassland over limestone | Grassland on deep soil over gypsum | Vegetation on shallow soil over gypsum | Vegetation on marl |
|-------------------|-------------|--------------------------|------------------------------------|----------------------------------------|--------------------|
| Number of species | 38* | 6 | 11 | 5 | 16* |
| P vs N | 0.50 | 0.88 | 0.68 | 0.18 | 0.45 |
| K vs N | 0.43 | 0.78 | 0.69 | 0.23 | 0.35 |
| Na vs N | 0.13 | 0.21 | 0.39 | 0.70 | 0.17 |
| Ca vs N | 0.08 | 0.89 | 0.19 | 0.10 | 0.14 |
| Mg vs N | 0.20 | 0.77 | 0.55 | 0.40 | 0.12 |
| N vs WCS | 0.07 | 0.86 | 0.16 | 0.57 | 0.03 |
| P vs WCS | 0.15 | 0.86 | 0.11 | 0.75 | 0.11 |
| K vs WCS | 0.14 | 0.71 | 0.28 | 0.19 | 0.30 |
| Na vs WCS | 0.44 | 0.16 | 0.25 | 0.87 | 0.80 |
| Ca vs WCS | 0.36 | 0.85 | 0.72 | 0.59 | 0.10 |
| Mg vs WCS | 0.18 | 0.86 | 0.02 | 0.18 | 0.30 |

*Number of species one fewer where P is involved.

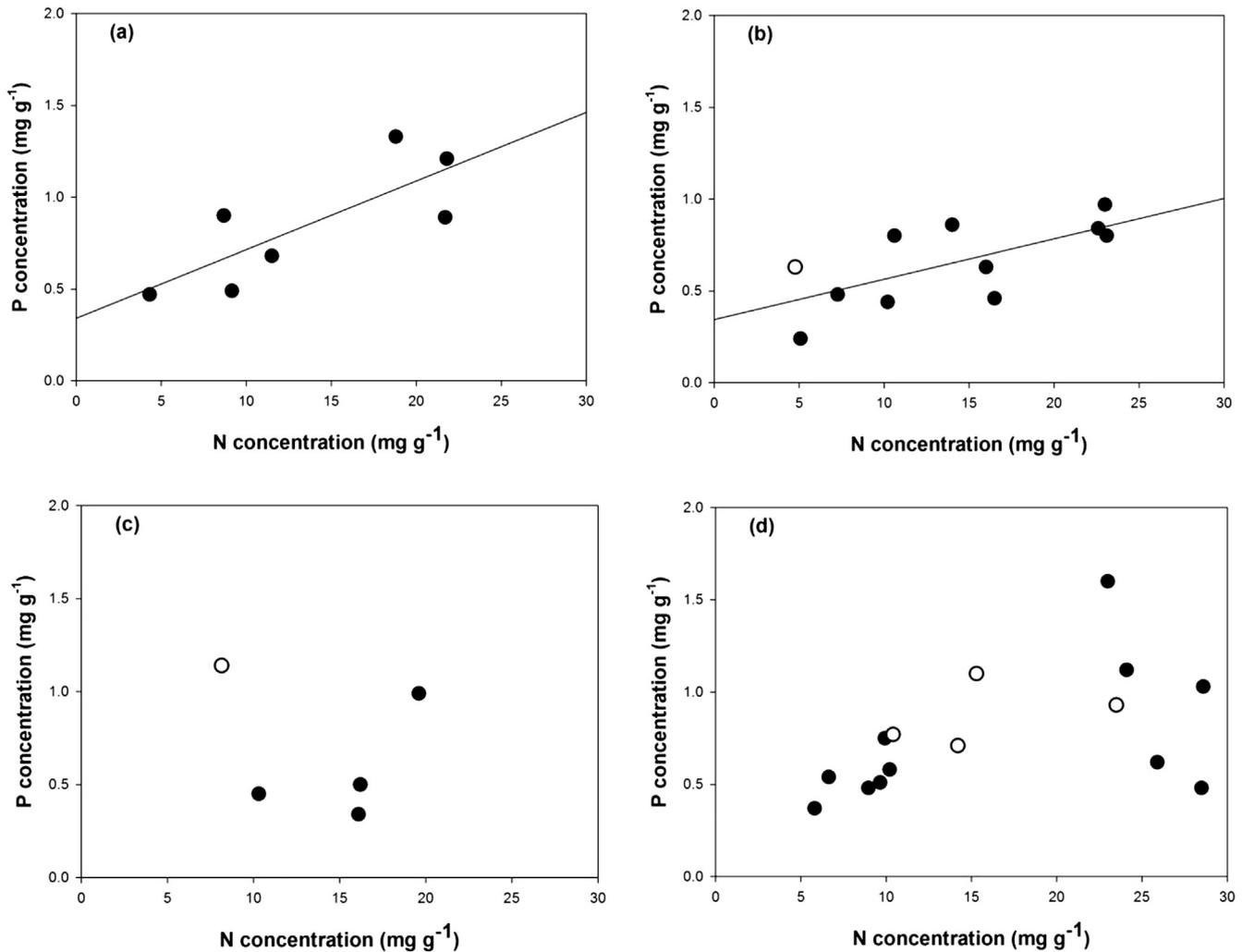


Fig. 5. The correlation of foliar nitrogen and phosphorus concentrations in different kinds of semi-desert: (a) grassland on limestone, and (b) grassland on deep soil over gypsum, (c) vegetation on shallow soil over gypsum, and (d) vegetation on marl. Empty circles indicate leaf-succulents, and filled circles other species.

The vegetation on marl differs significantly from that on limestone and the two types on gypsum in its high concentrations of K, Na and Mg. The higher mean [Na] is consistent with the higher concentration of Na in the soil solution, but the higher mean values for [K] and [Mg] are not paralleled in the concentrations of the exchangeable ions in the soil (Table 2).

3.10. Summary of results

The extent to which our expectations were met is shown in Table 6.

4. Discussion

4.1. Is there a wide range of N concentration among the different species within each type of semi-desert

Variation in [N] by a factor of about 5 was found in each of the three vegetation types with the greatest numbers of species analysed, and by a factor of 2.5–3.5 when grasses, highly succulent species, and species with shoots bearing scale leaves were excluded. Similarly, Carrick (2001) found variation by a factor of about 4 for 13 shrub species at one site in the karoo semi-desert of South Africa, representing a range of 8–35 mg g⁻¹, which is similar

that we found for southeastern Spain, if we lump shrubs from the three vegetation-types with most species sampled (7–34 mg g⁻¹).

After discussing the answers we obtained to the remaining questions set out in the Introduction we consider how far we can account for the wide range in [N] as associated with successional status or increased abundance on soils enriched by animals ('nitrophily').

4.2. How similar are the relationships between specific leaf area and foliar [N] across semi-arid vegetation types?

The correlation between SLA and foliar [N] was much stronger for non-succulent species in the grasslands on limestone and on deep soils over gypsum, and in open vegetation on shallow soils over gypsum, than in the whole collection of species in vegetation on marl (Fig. 2). Is variation in the degree of succulence responsible for the greater scatter shown by plants on marl? In the mid range of [N] in Fig. 2d two of the leaf-succulent species lay above the regression line (*Lycium intricatum* and *Euzomodendron bourgaeana*), while two lay below (*Suaeda* cf. *vera* and *Salsola oppositifolia*). This finding, coupled with the fact that *S. sediforme* fell on the regression line for all species in grassland on deeper soils over gypsum, while *S. gypsicola* was far above the line for other species on shallow soils over gypsum, emphasizes that no simple

Table 6
A summary of results.

| Expectation | Finding |
|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1. There will be a wide range of [N] in each vegetation type studied, possibly up to 5-fold | 5-fold range for grasslands on limestone and gypsum, and for vegetation on marl; range narrower for the smaller number of species on shallow soils over gypsum |
| 2. Within each vegetation type SLA will be correlated significantly with [N] | True for the two grasslands and for the non-succulent species on shallow soils over gypsum; for the vegetation on marl there was a marked but non-significant trend to higher SLA with higher [N] |
| 3. Within each vegetation type SLA will be correlated significantly with WCS, and there will be a significant correlation between [N] and WCS | True for the two grasslands and for the non-succulent species on shallow soils over gypsum; for the vegetation on marl there was a non-significant trend to a correlation between SLA and WCS, and a wedge-shaped relationship between WCS and [N] with a wider range of WCS at low [N] |
| 4. Among the perennial dicots SLA, [N] and WCS will be lower in species with longer-lived leaves | True for SLA and [N] but not WCS in the one balanced comparison within a vegetation type (4 species vs 5 in grassland on deep soils over gypsum); result same for all species lumped |
| 5. Among the perennial dicots SLA, [N] and WCS will be lower in species with larger leaves | Not true of any of the three variables; where there is an appreciable non-significant trend, it is in the reverse direction |
| 6. [P] will be correlated significantly with [N], but there will be a wide range in the N/P quotient | The correlation was found in only two of the vegetation types; N/P varied 2- to 5-fold within a vegetation type; the means for vegetation types varied from 14 to 26 |
| 7. WCS will be correlated with the concentrations on a dry mass basis of K and Mg (but not Na and Ca) in the vegetation on non-saline soils, and with the concentrations of Na and Mg in the vegetation on marl. Na will be the element accumulated in the greatest number of succulent and somewhat succulent species | Partly true; WCS was correlated with [Mg] and [Ca] but not [K] or [Na] in grassland on limestone; it was correlated with [Ca] but not [K], [Mg] or [Na] in grassland on deep soil over gypsum; and it was correlated with [Na] in plants on marl. No correlation was found for plants on shallow soil on gypsum. Na was accumulated in 12/16 species on marl; Mg accumulated in only four species, K and Ca in only two each; on shallow soil on gypsum two species accumulated Na |
| 8. The concentrations of nutrients in whole shoots will parallel those found in leaves alone; those of N and Mg (but not P, K, Ca and Na) will generally be lower in whole shoots than in leaves | Only partly true; the correlations were found as expected but the whole-shoot concentrations of N and Mg were not lower than those in leaves, while the concentrations of P and K were markedly lower than in leaves |
| 9. The mean foliar [Na] will be higher in the vegetation on marl than in the other vegetation | True but [K] and [Mg] were also higher in the vegetation in marl (though measures of availability in the soil were not higher), and [Ca] was higher in vegetation on shallow soil over gypsum |

generalization can yet be made about the SLA vs [N] relationship for leaf succulents relative to that for non-succulent species.

4.3. Are SLA and N correlated with WCS in the various types of vegetation, and does a more exact definition of 'succulence' help interpretation?

We found substantial variation among vegetation types in the relationships among SLA, N and WCS, and in several cases the relationships changed when succulents were excluded. However, distinguishing succulents rigorously requires a physiological point of view. Thus, the best definition of succulence involves more than a simple measure of the amount of water per unit dry mass or per unit area of leaf; rather, one should consider the amount of water per dry mass or area that can be stored and then used without passing the point where the leaf dies of desiccation, i.e. the 'available' water (Bartlett et al., 2012). The effectiveness of the 'available' water store will depend on the rate of loss, and thus the degree of desiccation before the stomata shut, the permeability of the cuticle, the surface to volume ratio, and the degree of self-shading within the shoot which affects the irradiance received by a leaf, and therefore the leaf temperature and the gradient of water potential from the leaf to the air. However, there is no published information on the amount of 'available' water in the leaf, or the rate of loss of water during a prolonged dry spell, for any of the species we studied. For the moment therefore we are bound to consider the two variables most often advocated in the literature as measures of 'succulence': the 'degree of succulence' (DOS, mass of water per unit area of leaf) proposed by Delf (1911), renamed 'water mass per unit area' (WMA) by Bartlett et al. (2012), and WCS, called 'saturated water content' (SWC) by Bartlett et al. (2012) and Ogburn and Edwards (2012).

Among the species we sampled there was a clear discontinuity in WCS between those categorized by taxonomists as leaf succulents and the rest. The four perennial leaf succulents we sampled on marl, the single annual we sampled on marl, and the two *Sedum* species we sampled on gypsum all had WCS values $>5 \text{ g g}^{-1}$, while

the rest all had lower values (Fig. 6a). There was an almost equally clear distinction between the leaf succulents and the rest using the WMA, but there was overlap between the taxonomists' leaf succulents and the scleromorphic grass *Lygeum spartum*. The latter had only a moderate WCS (2.6 g g^{-1}) but a high dry mass per unit leaf area (30.3 mg cm^{-2}), and thus a higher WMA (79 mg cm^{-2}) than the lowest value for an unquestioned leaf succulent (73 mg cm^{-2} for *Euzomodendron bourgaeum*). The result for *Lygeum* leads to the more general point that among the species we studied those not rated leaf succulent by the taxonomists showed no correlation between WMA and WCS ($r = 0.01$; Fig. 6b). For these plants only the WCS is a useful measure of succulence. However, in the broader context WCS by itself is of limited value in defining 'succulence' as ordinarily understood by a botanist, despite its being advocated by Ogburn and Edwards (2012). In the temperate zones particularly there are many species with thin soft leaves, a high SLA (c. $200\text{--}400 \text{ cm}^2 \text{ g}^{-1}$) and a high WCS (c. 5 g g^{-1}); they certainly do not have "succulent physiology". Good examples are the species of *Oxalis*, *Taraxacum* and *Trifolium* studied by Vendramini et al. (2002); their WMA values are low (in the range $11\text{--}22 \text{ mg cm}^{-2}$), close to the value of 10 mg cm^{-2} for *Mercurialis annua* which Delf (1911) used as a non-succulent comparator. We conclude that given the continuous spectra of values for both WCS and WMA, it may be helpful to set arbitrary criteria for the definition of a 'succulent' and to use a combination of WCS and WMA in the definition, e.g. $\text{WCS} > 5 \text{ g g}^{-1}$ and $\text{WMA} > 60 \text{ mg cm}^{-2}$. Further work is needed to see how far water storage occurs in distinct colourless tissues rather than in the chlorenchyma of the leaves of the various genera involved.

In the succulent-rich vegetation on marl the correlation between WCS and SLA was weak and not significant (Fig. 3d; $r = 0.24$). In the sample of 13 'leaf succulents' in western Argentina studied by Vendramini et al. (2002) – a mixture of native and introduced species – there was a significant relationship between WCS and SLA ($r = 0.72$; $P = 0.005$), but this was strongly dependent on the two species with the highest WCS values. Their 'leaf succulents' resemble the whole range of species that we studied on marl, plus

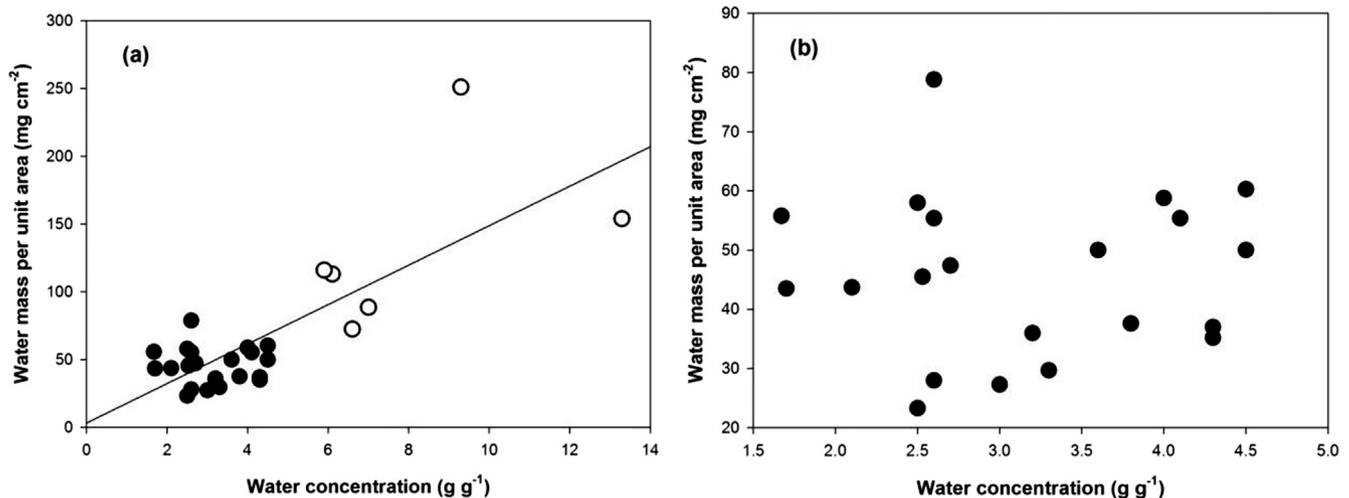


Fig. 6. The relationship between water mass per unit area (WMA) and water concentration at saturation (WCS) in (a) the total collection of species studied in the four vegetation types, and (b) those with $WCS < 5 \text{ g g}^{-1}$. Empty circles indicate leaf-succulents, and filled circles other species.

the two *Sedum* spp. sampled over gypsum, in that the WCS ranged from 1.7 to 19 g g^{-1} (cf. our range $1.6\text{--}13.3 \text{ g g}^{-1}$), and only 5/13 had $WCS > 5 \text{ g g}^{-1}$. Their 8 species with $WCS < 5 \text{ g g}^{-1}$ showed no appreciable trend to greater SLA with greater WCS ($r = 0.022$). We conclude that in general SLA is not correlated with WCS in succulents as it is in non-succulents.

The stem succulents in our study had lower WCS values than the leaf succulents ($2.5\text{--}2.7 \text{ g g}^{-1}$ for whole shoots including scale leaves; Table 4). New data on the WCS values of non-succulent stems are needed to provide comparison.

The relationship between [N] and WCS in succulent plants is discussed further in Appendix 3.

4.4. Do species with longer-lived leaves have lower SLA and/or lower [N] or WCS?

In our collection of species those with shorter-lived leaves had significantly higher [N] than those with longer-lived leaves, as found by earlier researchers in many kinds of vegetation (Wright et al., 2004a, 2004b). There was also a marked but non-significant trend to higher SLA ($P = 0.09$), but there was no appreciable trend in WCS. Notably, our classification into shorter- vs longer-lived leaves is a simple and preliminary approach; quantitative data for leaf longevity would provide a stronger test of the relationship with SLA.

The simplest interpretation is that the species with shorter-lived leaves have higher [N] and SLA because they invest less in generalized defence against physical and biological hazards (Grubb, 1986; Edwards, 1989), which means less investment in fibrous tissue and cell wall generally (John et al., 2013). Future studies are needed to resolve the basis in tissue anatomy and composition for the observed differences in [N] and SLA among our study species.

4.5. Is SLA correlated with leaf area in any of the types of vegetation?

The significant negative relationship between SLA and leaf area often found for plants of one life-form in a single vegetation-type was not found in any of the semi-arid vegetation types we studied. Where the relationship is significant, it likely requires a large enough range in leaf size for structural investment to be substantial in the larger leaves. An analogous effect is seen in the low SLA (and

low [N] and WCS) in the leaves of the tall grass *Stipa tenacissima* (Table 4); the individual leaf laminae are relatively narrow (up to 5 mm wide; FIP, personal observations) and very long (up to 70 cm; Blanca et al., 2011) and the leaf tissues have a stem-like function of support in keeping the leaf semi-upright (Valladares and Pugnaire, 1999). The semi-desert of SE Spain lacks species with large, thick succulent leaves either having minimal development of thick-walled outer tissues (as seen in some Aizoaceae-Mesembryanthemoideae) or having tough outer tissues with thick cell walls (as seen in *Agave* and *Aloe* species); both thick leaf-types can certainly have notably low SLA values (von Willert et al., 1990; Vendramini et al., 2002).

4.6. Are [N] and [P] correlated, and what is the range of N/P quotients in each of the vegetation types?

A significant correlation between foliar [N] and [P] was found for the grassland on limestone and for that on deep soil over gypsum, with a much lower increase in [P] for a given increase in [N] in the latter case. For plants on marl there was a wedge-shaped relationship between [P] and [N] with a narrow range of [P] at low [N] and a wide range at high [N]. It will be interesting to see if this latter pattern appears in vegetation on salty soils in other semi-desert regions.

The N/P quotients vary widely within each of the communities studied, most strikingly among the species on marl (12–59) and in the grassland on deeper soils over gypsum (8–36), in both cases overlapping widely the critical range of 14–16 hypothesized by Koerselman and Meuleman (1996) to separate limitation by N (<14) and limitation by P (>16). Our mean values for the various vegetation types (14, 22, 23 and 26) also span across the critical range, much as the mean values in two studies in the Mojave Desert did (10 and 21; Table S1). If, despite the finding of Drenovsky and Richards (2004) in one critically investigated case, the N:P quotient is generally a useful guide to the primary limiting nutrient, then that nutrient will vary within and between the communities we studied. It is very desirable that a field experiment be set up to test the primary limiting nutrient(s) in contrasted communities in semi-arid Spain. It seems likely that the various species in a semi-desert community will respond differently to additions of N and P as was found by Drenovsky and Richards (2004) and as seen in the much earlier studies on mesic grassland by J.B. Laws & J.H. Gilbert in

1880 (summarized by Thurston, 1969) and on the herb layer in a temperate deciduous forest by W. Grabherr in 1942 (summarized by Ellenberg, 1988).

Variation in N/P quotient may reflect differences in allocation to cell walls, which have higher [N] relative to [P] than protoplasts, and/or may be inversely related to cold tolerance and/or growth rate (Niinemets et al., 1999; Shipley et al., 2006; Elser et al., 2010; Waite and Sack, 2011).

4.7. Are the concentrations of any of the basic cations correlated with WCS, and which basic cations are accumulated in the succulent species?

As with the variables SLA, [N] and WCS, the simplest case is that of the grassland on limestone. WCS was significantly correlated with [Ca] and [Mg] (Table 5), and marginally with [K] ($P = 0.072$). Insofar as these cations and the cell water are located chiefly in the vacuole rather than the cell wall or cytoplasm the species in different families are maintaining much the same vacuolar concentrations irrespective of the WCS. The species in grassland on deeper soils over gypsum showed this pattern only for [Ca], and for plants on shallow soils over gypsum there was no significant correlation between WCS and the concentration of any major basic cation. The range of WCS was much greater for plants on marl; it was correlated with [Na] but not [K], [Ca] or [Mg] (Table 5). Clearly the plants which accumulated the Na to high levels were also the succulent ones, accumulating more water per unit dry mass.

4.8. Concentrations in whole shoots versus leaves

The concentrations in whole shoots were generally close to those found for fully expanded leaves alone (Appendix Table S2) with the exception of K which had values about half those in leaves. The differences in K should be taken into account if leaf values are used to estimate the browsing nutrient intake by browsers. In semi-arid systems shoot chemical composition varies with phenology and differences in rainfall, which influence the browsing intensity by large herbivores. In general browsers prefer young shoots and new leaves with higher protein and lower total fibre contents (Watson and Owen-Smith, 2002).

Whole shoots of *Salsola oppositifolia* from the North African Chafarinas Islands analysed by García et al. (2002) had similar nutrient concentrations to those found in semi-arid SE Spain. However, at the island sites affected by seabird guano the shoot concentration of *Salsola* significantly increased for N (x 1.9), P (x 4.0) and K (x 1.8) but decreased for Ca (x 1.7) in association with poorer performance and toxicity symptoms; ultimately the species was replaced by guano-tolerant *Suaeda vera* (García et al., 2002).

4.9. Differences among vegetation types in mean foliar concentrations, and possible relationship to differences in soil chemistry

The most striking feature of the differences in foliar elemental concentrations between vegetation types is the extent to which inherent differences between species dominate over the variation resulting directly from different concentrations of the relevant ions in the soils.

We found no relationship between the mean foliar [N] of a vegetation type and the total [N] in the soil. The four soils on marl had markedly lower [N] than three of the four soils over limestone and both of the deep soils over gypsum but the plants of the vegetation on marl did not have lower foliar [N]: soils 0.05–0.08 vs (0.15–)1.3–2.3 and 0.31 (both sites) mg g⁻¹ (Table 2); leaves 17.5 ± 2.25 vs 12.4 ± 2.71 and 13.9 ± 2.10 (Appendix Table S3).

Neither of the two species collected on both marl and limestone had a lower foliar [N] on marl: *Stipa tenacissima* 8.0 vs 4.3 mg g⁻¹, and *Thymelaea hirsuta* 9.9 vs 8.7 mg g⁻¹ (Table 4). Similarly neither of the two species sampled on both marl and deep soil over gypsum had a lower foliar [N] on marl: *Anthyllis terniflora* 29 vs 23 mg g⁻¹, and *Hammada articulata* 23 mg g⁻¹ on both soil-types (Table 4). We discuss the evidence for P and cations in Appendix 3.

Examples of phylogenetic effects on differences in foliar nutrient concentrations among nearby vegetation types have long been known, e.g. in the study by Tanner (1977) of adjacent montane rain forests on different soil types in Jamaica, but the incidence of such effects has become increasingly clear for both tropical and temperate vegetation over the last two decades (Metali et al., 2015).

4.10. Interpretation of the wide range of foliar [N] and associated properties in a given vegetation type

We have already seen that [N] is greater in shorter-lived leaves (4.4 above). In Appendix 3 we show that the species in the stands on marl that we studied are a mixture of those associated with N-enriched soils and those not so associated, and that surprisingly these did not differ significantly in mean [N].

We now consider whether or not the higher foliar [N] values of certain species in the grasslands are correlated with early-successional status. On a world scale the simplest interpretation of the parts played by the various species in the dynamics of communities is that those with higher SLA, foliar [N] and WCS are more dependent on disturbance and found earlier in succession. We accept that this is often found, but there are certainly marked exceptions, notably certain conifers (species of *Juniperus* and *Pinus*) which invade relatively early in succession in Northern Temperate areas where the late-successional vegetation is deciduous, but have relatively long-lived leaves with low [N].

Among the non-succulent species we studied *Anthyllis cytioides*, *A. terniflora* and *Artemisia barrelieri* conform to the paradigm. All three species were noted by Freitag (1971a) and/or Peinado et al. (1992) as benefitting from disturbance which opens the canopy and presumably reduces the intensity of root competition locally. All three have shorter-lived leaves (Table 3) and relatively high SLA and [N] (Table 4). *Hammada articulata* is likewise invasive and the shoots are rich in N (Table 4), despite having minute leaves of unknown life-length. On the other hand the invasive *Thymelaea hirsuta* is functionally like an invasive species of *Pinus* with longer-lived leaves (Table 3) and a low N concentration (Table 4).

It remains the case that there is wide variation in foliar [N], when we leave aside species noted as benefitting from disturbance, and avoid the complications of grasses which provide stem-like support in their leaves, and species with scale leaves. It seems likely that in general a wide range in [N] and associated properties are found in plants that fill much the same role in a given community, as argued by Grubb (2002) for a collection of strongly shade-tolerant tree species in Australian tropical lowland rain forest. On limestone the range is from 9.2 (*Rosmarinus officinalis*) to 18.8 (*Phlomis purpurea*). On deep soils over gypsum it is from 7.3 (*Rosmarinus eriocalyx*) to 16.5 (*C. hispanica*), and on shallow soils on gypsum it is from 10.3 (*Teucrium turredanum*) to 19.6 (*S. viscosa*).

5. Conclusions and way forward

The interrelationships between the leaf properties we studied mostly accord with those established in other kinds of vegetation around the world, especially for plants in the human-induced grasslands derived from various woody communities, and for the non-succulent plants on shallow soils over gypsum. However, for

the plants on marl, where there are many succulents, the inter-relationships between important leaf properties are not as in non-succulent vegetation. New work is needed to understand why the inter-relationships are different, preferably coupled with work in a species-rich semi-desert region with a different flora, e.g. the Karoo in South Africa. For plants in all the vegetation types studied new work is needed to establish more precisely the relationship between leaf properties and leaf longevity, to relate the rates of net assimilation to leaf structure and chemistry, and to check on the annual cycle of changes in leaf properties.

Experiments are needed in each of the vegetation-types studied to test whether the foliar N/P quotient is more often than not an effective indicator of the primary limiting nutrient for a given species. The wide range in that quotient makes the vegetation of the study area highly suitable for testing the ideas put forward by Koerselman and Meuleman (1996). The existence of a wide range of [N] in one tightly defined group of species, e.g. late-successional shrubs in semi-desert on either limestone or marl, remains a puzzle. New work is needed to explore the advantages and disadvantages for plants of one functional type having a given foliar [N] with its associated values for leaf longevity and net assimilation.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.jaridenv.2015.03.001>.

References

- Armas, C., Miranda, J.D., Padilla, F.M., Pugnaire, F.I., 2011. The Iberian Southeast. Preface. *J. Arid Environ.* 75, 1310–1320.
- Bartlett, M.K., Scoffoni, C., Sack, L., 2012. The determinants of leaf turgor loss point and prediction of drought tolerance of species and biomes: a global meta-analysis. *Ecol. Lett.* 15, 393–405.
- Beadle, N.C.W., 1981. *The Vegetation of Australia*. Cambridge University Press, Cambridge, UK.
- Blanca, G., Cabezudo, B., Cueto, M., Salazar, C., Morales Torres, C. (Eds.), 2011. *Flora Vascular de Andalucía Oriental*, second ed. Universidades de Almería, Granada, Jaén and Málaga, Granada.
- Bornkamm, R., Kehl, H., 1990. The plant communities of the Western Desert of Egypt. *Phytocoenologia* 19, 149–231.
- Carrick, P.J., 2001. *Shrub Community Dynamics in a South African Semi-desert* (Ph.D. dissertation). University of Cambridge, UK.
- Delf, E.M., 1911. Transpiration and behaviour of stomata in halophytes. *Ann. Bot.* 25, 485–505.
- Drenovsky, R.E., Richards, J.H., 2004. Critical N: P values predicting nutrient deficiencies in desert shrublands. *Plant & Soil* 259, 59–69.
- Edwards, P.J., 1989. Insect herbivory and plant defence theory. In: Grubb, P.J., Whittaker, J.B. (Eds.), *Toward a More Exact Ecology*. Blackwell Scientific, Oxford, pp. 275–297.
- El-Ghonyemy, A.A., El-Gazaar, A.M., Wallace, A., Romney, E.M., 1977. Mineral element composition of the natural vegetation along a transect at Mareotis, Egypt. *Soil Sci.* 124, 16–26.
- Ellenberg, H., 1988. *Vegetation Ecology of Central Europe*, fourth ed. Cambridge University Press, Cambridge.
- Elser, J.J., Fagan, W.F., Kerkhoff, A.J., Swenson, N.G., Enquist, B.J., 2010. Biological stoichiometry of plant production: metabolism, scaling and ecological response to global change. *New Phytol.* 186, 593–608.
- Freitag, H., 1971a. Die natürliche vegetation des sudostspanischen Trockengebietes. *Bot. Jahrb.* 91, 147–308.
- García, L.V., Marañón, T., Ojeda, F., Clemente, L., Redondo, R., 2002. Seagull influence on soil properties, chenopod shrub distribution, and leaf nutrient status in semi-arid Mediterranean islands. *Oikos* 98, 75–86.
- Grove, A.T., Rackham, O., 2001. *The Nature of Mediterranean Europe: an Ecological History*. Yale University Press, New Haven.
- Grubb, P.J., 1977. The control of forest growth and distribution on wet tropical mountains: with special reference to mineral nutrition. *Annu. Rev. Ecol. Syst.* 8, 83–107.
- Grubb, P.J., 1986. Sclerophylls, pachyphylls and pycnophylls: the nature and significance of hard leaf surfaces. In: Juniper, B.E., Southwood, T.R.E. (Eds.), *Insects and the Plant Surface*. Arnold, London, pp. 141–165.
- Grubb, P.J., 1998. A reassessment of the strategies of plants which cope with shortages of resources. *Perspect. Plant Ecol. Evol. Syst.* 1, 3–31.
- Grubb, P.J., 2002. Leaf form and function – towards a radical new approach. *New Phytol.* 155, 317–320.
- Grubb, P.J., 2005. The impact of nutrient supply on the kind of vegetation found in the western semi-deserts of southern Africa. *Trans. R. Soc. S. Afr.* 59, 79–80.
- Grubb, P.J., Edwards, P.J., 1982. Studies of mineral cycling in a montane rain forest in new Guinea. III. The distribution of mineral elements in the above-ground material. *J. Ecol.* 70, 623–648.
- Grubb, P.J., Turner, I.M., Burslem, D.F.R.P., 1994. Mineral nutrient status of coastal hill dipterocarp forest and *Adinandra Belukar* in Singapore: analysis of soil, leaves and litter. *J. Trop. Ecol.* 10, 559–577.
- Güsewell, S., 2004. N: P ratios in terrestrial plants: variation and functional significance. *New Phytol.* 164, 243–266.
- John, G.P., Scoffoni, C., Sack, L., 2013. Allometry of cells and tissues within leaves. *Am. J. Bot.* 100, 1936–1948.
- Koerselman, W., Meuleman, A.F.M., 1996. The vegetation N: P ratio: a new tool to detect the nature of nutrient-limitation. *J. Appl. Ecol.* 33, 1441–1450.
- Lázaro, R., Rodrigo, F.S., Gutiérrez, L., Domingo, F., Puigdefábregas, J., 2001. Analysis of a 30-year rainfall record (1967–1997) in semi-arid SE Spain for implications on vegetation. *J. Arid Environ.* 48, 373–395.
- Marañón, T., Grubb, P.J., 1993. Physiological basis and ecological significance of the seed size-relative growth rate relationship in Mediterranean annuals. *Funct. Ecol.* 7, 591–599.
- Martínez, J.P., Ledent, J.F., Bajji, M., Kinet, J.M., Lutts, S., 2003. Effect of water stress on growth, Na⁺ and K⁺ accumulation and water use efficiency in relation to osmotic adjustment in two populations of *Atriplex halimus* L. *Plant Growth Regul.* 41, 63–73.
- Metali, F., Abu Salim, K., Tennakoon, K., Burslem, D.F.R.P., 2015. Controls on foliar nutrient and aluminium concentrations in a tropical tree flora: phylogeny, soil chemistry and interactions between elements. *New Phytol.* 205, 280–292.
- Milla, R., Reich, P.B., 2007. The scaling of leaf area and mass: the cost of light interception increases with leaf size. *Proc. R. Soc. B* 274, 2109–2114.
- Niinemets, U., Tenhunen, J.D., Canta, N.R., Chaves, N.R., Faria, T., Pereira, J.S., Reynolds, J.F., 1999. Interactive effects of nitrogen and phosphorus on the acclimation potential of foliar photosynthetic properties of cork oak, *Quercus suber*, to elevated atmospheric CO₂ concentrations. *Glob. Change Biol.* 5, 455–470.
- Niklas, K.J., Cobb, E.D., Niinemets, U., Reich, P.B., Sellin, A., Shipley, B., Wright, I.J., 2007. “Diminishing returns” in the scaling of functional leaf traits across and within species groups. *Proc. Natl. Acad. Sci. U. S. A.* 104, 8891–8896.
- Nobel, P.S., Quero, E., Linares, H., 1988. Differential growth responses of agaves to nitrogen, phosphorus, potassium and boron applications. *J. Plant Nutr.* 11, 1683–1700.
- Ogburn, R.M., Edwards, E.J., 2012. Quantifying succulence: a rapid physiologically meaningful metric of plant water storage. *Plant Cell Environ.* 35, 1533–1542.
- Osnas, J.L., Lichstein, J.W., Reich, P.B., Pacala, S.W., 2013. Global leaf trait relationships: mass, area, and the leaf economics spectrum. *Science* 340, 741–744.
- Peinado, M., Alcaraz, F., Martínez-Parras, J.M., 1992. *Vegetation of South-eastern Spain*. Cramer, Berlin.
- Penning de Vries, F., Krul, J., van Keulen, H., 1980. Productivity of Sahelian rangelands in relation to the availability of nitrogen and phosphorus from the soil. In: Rosswall, T. (Ed.), *Nitrogen Cycling in West African Ecosystems*. Royal Swedish Academy of Science, Stockholm, pp. 95–103.
- Reich, P.B., Ellsworth, D.S., Walters, M.B., Vose, J.M., Gresham, C., Volin, J.C., Bowman, W.D., 1999. Generality of leaf trait relationships: a test across six biomes. *Ecology* 80, 1955–1969.
- Reich, P.B., Walters, M.B., Ellsworth, D.S., 1997. From tropics to tundra: global convergence in plant functioning. *Proc. Natl. Acad. Sci. U. S. A.* 94, 13730–13734.
- Roderick, M.L., Berry, S.L., Saunders, A.R., Noble, I.R., 2000. On the relationship between the composition, morphology and function of leaves. *Funct. Ecol.* 13, 696–710.
- Shaltout, K.H., 1992. Nutrient status of *Thymelaea hirsuta* (L.) Endl. in Egypt. *J. Arid Environ.* 23, 423–432.
- Shipley, B., 1995. Structured interspecific determinants of specific leaf area in 34 species of herbaceous angiosperms. *Funct. Ecol.* 9, 312–319.
- Shipley, B., Lechowicz, M.J., Wright, I., Reich, P.B., 2006. Fundamental trade-offs generating the worldwide leaf economics spectrum. *Ecology* 87, 535–541.
- Sokal, R.R., Rohlf, F.J., 1987. *Introduction to Biostatistics*. Freeman & Co, San Francisco, USA.
- Specht, R.L., 1972. *The Vegetation of South Australia*, second ed. Government Printer, Adelaide, Australia.
- Tanner, E.V.J., 1977. Four montane rain forests in Jamaica: a quantitative characterization of the floristics, the soils and foliar nutrient levels, and a discussion of the interrelations. *J. Ecol.* 65, 883–918.

- Thurston, J.M., 1969. The effect of liming and fertilizers on the botanical composition of permanent grassland, and on the yield of hay. In: Rorison, I.H. (Ed.), *Ecological Aspects of the Mineral Nutrition of Plants*. Blackwell Scientific Publications, Oxford, pp. 3–10.
- Turner, I.M., 1994. Sclerophylly: primarily protective? *Funct. Ecol.* 8, 669–675.
- Valladares, F., Pugnaire, F.I., 1999. Tradeoffs between irradiance capture and avoidance in semi-arid environments assessed with a crown architecture model. *Ann. Bot.* 83, 459–469.
- van de Veg, M.E., Mair, P., Grace, J., Atkin, O.K., 2009. Altitudinal variation in leaf mass per unit area, leaf tissue density and foliar nitrogen and phosphorus content along an Amazon-Andes gradient in Peru. *Plant Ecol. Divers.* 2, 243–254.
- Vendramini, F., Diaz, S., Gurvich, D.E., Wilson, P.J., Thompson, K., Hodgson, J.G., 2002. Leaf traits as indicators of resource-use strategy in floras with succulent species. *New Phytol.* 154, 147–157.
- von Willert, D.J., Eller, B.M., Werger, M.J.A., Brinckmann, E., 1990. Desert succulents and their life strategies. *Vegetatio* 90, 133–143.
- Waite, M., Sack, L., 2011. Does global stoichiometry theory apply to bryophytes? tests across an elevation x soil age ecosystem matrix on Mauna Loa, Hawaii. *J. Ecol.* 99, 122–134.
- Walter, H., 1964. Die Vegetation der Erde in öko-physiologischer Betrachtung. In: *Die tropischen und subtropischen Zonen*, vol. I. Fischer, Jena, Germany.
- Watson, L.H., Owen-Smith, N., 2002. Phenological influences on the utilization of woody plants by eland in semi-arid shrubland. *Afr. J. Ecol.* 40, 65–75.
- Wilson, P.J., Thompson, K., Hodgson, J.G., 1999. Specific leaf area and leaf dry matter content as alternative predictors of plant strategies. *New Phytol.* 143, 155–162.
- Wright, I.J., Westoby, M., 2002. Leaves at low versus high rainfall: coordination of structure, lifespan and physiology. *New Phytol.* 155, 403–416.
- Wright, I.J., et al., 2004a. The worldwide leaf economics spectrum. *Nature* 428, 821–827.
- Wright, I.J., Groom, P.K., Lamont, B.B., Poot, P., Prior, L.D., Reich, P.B., Schulze, E.-D., Veneklaas, E.J., Westoby, M., 2004b. Leaf trait relationships in Australian plant species. *Funct. Plant Biol.* 31, 551–558.
- Yue, L.J., Li, S.X., Ma, Q., Zhou, X.R., Wu, G.Q., Bao, A.K., Zhang, J.L., Wang, S.M., 2012. NaCl stimulates growth and alleviates water stress in the xerophyte *Zygo-phyllum xanthoxylum*. *J. Arid Environ.* 87, 153–160.