Co-variations in litter decomposition, leaf traits and plant growth in species from a Mediterranean old-field succession

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Summary

1. A growing consensus is developing that the impact of species on ecosystem properties is mediated, at least partially, by the traits of their component species. A previous study demonstrated that the field decomposition of complex litters produced by different communities of a Mediterranean successional sere was related to the average trait value of these communities. Here we scale down to the species level, to test whether similar relationships are found for selected species from these communities. We also test whether litter decomposability can be considered as part of the suite of traits characterizing the fast–slow growth continuum in plants.

2. We chose 12 of the most abundant herbaceous species characteristics of three stages of the old-field succession mentioned above. We investigated trait variation and covariation for the eight following traits: specific leaf area (SLA), leaf phosphorus (LPC), nitrogen (LNC) and carbon (LCC) concentrations, leaf dry matter content (LDMC) and leaf total phenols (TPh), all on material collected in the field; and litter decomposability ($K_{pot}$) and maximum relative growth rate ($RGR_{max}$), obtained under standardized conditions in the laboratory.

3. Five of these traits were significantly lower in species from the advanced successional stage. These trends were similar when comparisons were conducted either with the 12 species, or on a subset incorporating taxonomic information. LDMC was the single trait best correlated with species $RGR_{max}$ and $K_{pot}$; the two latter traits were also significantly correlated with one another.

4. These results provide clear evidence of functional links between plant growth, leaf traits and litter decomposability. LDMC appears as a pivotal trait of living leaves related to their structural properties. It influences the quality of the litter produced, and hence species’ potential ‘after-life effects’ on ecosystem properties.

Key-words: Leaf traits, litter decomposability, relative growth rate, secondary succession

Introduction

Land-use change is considered as one of the main anthropogenic drivers leading to modifications in the composition and functioning of ecosystems (Vitousek et al. 1997). A growing consensus is currently developing that the impacts of such changes on ecosystem properties should be attributed, at least partially, to changes in species’ functional traits (Chapin et al. 2000; Diaz & Cabido 2001; Lavorel & Garnier 2002; Eviner & Chapin 2003). In the case of land abandonment in the Mediterranean region of Southern France (Debussche et al. 1999), a previous study has shown that changes in several ecosystem properties during the course of secondary succession were related to changes in plant traits (Garnier et al. 2004). Species with attributes allowing a rapid acquisition of resources were progressively replaced by species with attributes allowing efficient conservation of resources; this was related to an increase in standing biomass and carbon stocks, but a decrease in ecosystem process rates such as specific primary productivity and litter decomposition rate. Here we focus on the latter, with
the aim of scaling down to the species level, and test whether the decomposability of representative species from the same secondary succession also decreases with increasing age of abandonment.

Several studies have shown that leaf litter decomposability relates to the physical properties of living organs such as leaf toughness (Cornelissen et al. 1999; Pérez-Harguindeguy et al. 2000) and chemical composition (reviewed by Wardle 2002). In the successional sere described above, ecosystem-level decomposition of litter was correlated with community-aggregated (weighted according to relative abundance of species) values of specific leaf area (SLA), leaf dry matter content (LDMC) and leaf nitrogen concentration (LNC) (Garnier et al. 2004). We therefore tested whether these traits of living leaves are also related to the litter decomposability of individual species. Several other traits known to affect decomposition were incorporated in this study: leaf phosphorus, carbon and phenol concentrations (Aerts & de Caluwe 1997; Wardle 2002).

SLA, LNC and LDMC, which are all related to the ecosystem-level decomposition rate, are part of the ‘economics spectrum’ described by Wright et al. (2004) at the leaf level, and are tightly related to a species’ potential relative growth rate (RGR_{max}) (Poorter & Garnier 1999). Limited evidence suggests a direct relationship between species’ litter decomposability and RGR_{max} in woody species (Cornelissen et al. 1999), but whether this trend is also valid in herbaceous species remains an open question (Cornelissen & Thompson 1997). This was also examined in the present study. Finally, how to relate the decomposition data obtained at the species level to the decomposition rate of complex communities is briefly discussed.

The questions raised above were addressed using 12 of the most abundant species of the old-field succession studied by Garnier et al. (2004).

Materials and methods

STUDY SITE AND SPECIES SELECTION

The study area is located in the Mediterranean region of southern France (43°51’ N, 3°56’ E, 100–160 m asl). In this region the climate is Mediterranean subhumid (Daget 1977), with a marked summer drought, frequent frosts in winter and unpredictability of precipitation in timing and amount, with generally frequent heavy rainfall events in autumn. Species were collected in old fields that were abandoned 2–45 years prior to our study, following removal of vines. A detailed description of the sites is given by Garnier et al. (2004). Herbaceous species were dominant in all the fields.

Twelve herbaceous species were selected among the most abundant species occurring in this successional sere, based on their abundance in these sites and their usual position in Mediterranean old-field successions (Escarré et al. 1983; Garnier et al. 2004). Three main stages were recognized based on the time since vineyard abandonment: early (2–4 years, median = 3); intermediate (7–15 years, median = 10); and advanced (15–45 years, median = 25). Four species were chosen for each of the three successional stages (Fig. 1).

Among these, five species pairs were chosen to form phylogenetically independent contrasts (PICs). Such analyses explicitly recognize that species share many characteristics as a consequence of their common ancestry. Our intention was to test whether there were shifts that go in a similar direction for different lineages as succession proceeds. Two criteria were used to construct these PICs: first, each species pair shared a more recent common ancestor than any other species (Soltis et al. 2000), thus each species pair represented a separate evolutionary divergence. Second, in each PIC, species belonging to the more contrasting stages (early vs advanced) were chosen when possible. Most PICs were species within the same family, but one contrast was chosen at a higher taxonomic level (Fig. 1).

COLLECTION OF MATERIAL

Traits of green leaves were measured on 10 replicate samples per species collected in the fields described above during spring 2002, on the youngest fully expanded, well lit leaves at the spring peak of growth. For all species these corresponded to 10 different individuals. Stems or twigs bearing healthy leaves were severed from a plant, rapidly wrapped in moist paper, placed in plastic bags and stored in a cool box until further processing in the laboratory (see Garnier et al. 2001 for methods).

Leaf litter was collected at the season of maximum leaf senescence for each species. In species that shed their litter (e.g. Sanguisorba minor) we collected dead leaves that dropped after gently shaking plants. In species that retain dead leaves on the plant (e.g. Brachypodium phoenicooides, Dactylis glomerata) or that die back completely above ground (Avena barbata, Bromus madritensis, Crepis foetida) we cut off leaves that were dead. Litters were carefully cleaned, then air-dried and stored in the laboratory.

GREEN LEAVES: STRUCTURE AND CHEMISTRY

Five leaf traits were measured on each species using standardized procedures (Cornelissen et al. 2003). Water-saturated specific leaf area (SLA_{sat}) and leaf dry matter content (LDMC_{sat}) (see Table 1 for abbreviations) were calculated as the ratio between leaf area and leaf dry mass, and between leaf dry mass and saturated fresh mass, respectively. For each species, two or three leaf samples were subsequently pooled together to obtain three to five batches (four in most cases) for nitrogen and carbon analyses. These bulked samples were then ground individually, and their total nitrogen (LNC) and carbon (LCC) concentration determined with an elemental analyser (Carlo Erba Instruments, model
atpB (Soltis et al. 2000), E (early), I (intermediate) and A (advanced) denote the successional stage in which these species most commonly occur. PIC = phylogenetically independent contrast.

Table 1. List of traits, abbreviations and units

<table>
<thead>
<tr>
<th>Trait</th>
<th>Abbreviation</th>
<th>Unit</th>
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<tbody>
<tr>
<td>Specific leaf area</td>
<td>SLA&lt;sub&gt;sat&lt;/sub&gt;</td>
<td>m&lt;sup&gt;2&lt;/sup&gt; kg&lt;sup&gt;-1&lt;/sup&gt;</td>
</tr>
<tr>
<td>Leaf dry matter content</td>
<td>LDMC&lt;sub&gt;sat&lt;/sub&gt;</td>
<td>mg g&lt;sup&gt;-1&lt;/sup&gt;</td>
</tr>
<tr>
<td>Leaf nitrogen concentration</td>
<td>LNC</td>
<td>mg g&lt;sup&gt;-1&lt;/sup&gt;</td>
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<tr>
<td>Leaf carbon concentration</td>
<td>LCC</td>
<td>mg g&lt;sup&gt;-1&lt;/sup&gt;</td>
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<tr>
<td>Leaf phosphorus concentration</td>
<td>LPC</td>
<td>mg g&lt;sup&gt;-1&lt;/sup&gt;</td>
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<tr>
<td>Total soluble phenol</td>
<td>TPh</td>
<td>mg g&lt;sup&gt;-1&lt;/sup&gt;</td>
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<tr>
<td>Potential relative growth rate</td>
<td>RGR&lt;sub&gt;max&lt;/sub&gt;</td>
<td>g g&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;</td>
</tr>
<tr>
<td>Potential decomposability</td>
<td>K&lt;sub&gt;pot&lt;/sub&gt;</td>
<td>g kg&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;</td>
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*mg gallic acid equivalent g<sup>-1</sup>.

EA 1108, Milan, Italy). Leaf phosphorus concentration (LPC) was determined on leaf material harvested at the same time as leaves for SLA<sub>sat</sub> determination, and presenting similar characteristics. Four replicates, consisting of ~20 mg leaf material, were ground and digested using concentrated sulphuric acid and hydrogen peroxide at 100 °C for 35 min and 360 °C for 2 h. Phosphorus concentration of these digests was measured colorimetrically with an autoanalyser (Alliance Instruments, Evolution II, Frépillon, France) using the molybdenum blue method (Grimshaw et al. 1989).

Finally, to measure total phenols, four samples of leaves (500 mg) or litter (1 g, dry mass) of each species were extracted twice (2 × 100 ml) under reflux into 50% (v/v) boiling ethanol for 30 min. After combination, these extracts were evaporated in a rotating vacuum evaporator, and the residues were dissolved in a standard volume of hot distilled water. Total phenol determinations were made from aqueous solutions with Folin--Ciocalteus reagent, following the method of Marigo (1973) with gallic acid used as a standard.

**Litter decomposition**

The litter was incubated in microcosms in the laboratory, under controlled conditions of temperature and humidity. The microcosms were of the type described by Taylor & Parkinson (1988). One kilogram of a previously prepared soil mixture consisting of mineral soil and surface organic horizon (3 : 1) from a nearby field was placed on a grid situated 2 cm above the bottom of each microcosm. The litter, previously soaked for 24 h in 0·1 l water, was enclosed in a thin litter bag of 0·3 mm nylon mesh (Northern Mesh, Oldham, UK) to recover all the material at each sampling time, and placed on the soil in the microcosms. In order to keep all the soluble nutrients in the system, the soaking water was poured into the microcosm, and the quantity of additional water needed to bring the water content of the microcosm soils up to 80% field capacity was calculated by weighing. To maintain constant soil moisture during incubation, microcosms were weighed each week and the quantity of water needed to replace that evaporating was added. Microcosms were maintained in the dark at 22 °C throughout the experiment.

For each of the 12 species, 10 samples (3·00 ± 0·01 g each) were weighed and placed in microcosms for incubation. Two replicate samples per date were removed from the microcosms at 1, 2, 4, 6 and 8 weeks. Soil particles were removed from the litter bags, and each sample was weighed after drying in a fan oven for 48 h at 60 °C (see Gillon et al. 1999 for details). Three additional samples of each species were weighed after drying in an oven at 60 °C for 48 h to determine the exact water content of the original litter.

The percentage of oven-dried litter mass remaining is denoted %MR. The single negative exponential model proposed by Olson (1963) gave the best fit to the %MR values for each species: %MR = 100e<sup>-Kt</sup>, where K (= K<sub>pot</sub>) is the decomposition rate constant over time t in days; and %MR is expressed as a percentage of the original mass. The significance of the K<sub>pot</sub> value was tested using a t-test and species-specific differences were tested with ANOVA (see Data analysis).

**Growth analysis**

To determine the RGR<sub>max</sub> of seedlings, seeds from naturally occurring plants were collected in the study area in 2002. Seeds were germinated in a growth room in compartmented plastic trays filled with moist silica. After germination, seedlings were transferred to the growth chamber as soon as they were sufficiently large...
to be manipulated without damage. This transplantation procedure ensured minimum size/age variation between seedlings.

RGR$_{\text{max}}$ of hydroponically grown seedlings was obtained for all species except Rubia peregrina, seeds of which did not germinate. The experiment was set up in a growth chamber in which metal halide and high-pressure sodium lamps provided light (500 µmol m$^{-2}$ s$^{-1}$) for 15-5 h day$^{-1}$, while temperatures were 24 °C during the light period and 19 °C during the dark. Relative humidity was > 70%. The nutrient solution consisted of 2 mM KNO$_3$, 1·5 mM Cu(NO$_3$)$_2$·4H$_2$O, 2 mM Mg(SO$_4$)$_2$·7H$_2$O, 0·5 mM (NH$_4$)$_2$SO$_4$, 1 mM KH$_2$PO$_4$, 0·01 mM MnSO$_4$·H$_2$O, 1 µM Na$_2$MoO$_4$·2H$_2$O, 46 µM H$_3$BO$_3$, 1 µM ZnSO$_4$·7H$_2$O, 1 µM CuSO$_4$, 68·1 µM EDTA-Fe. The pH was measured daily and adjusted to 5·8. The nutrient solution was continually oxygenated with aquarium air pumps and mixed with submerged pumps. The solution was changed weekly or when nitrate levels decreased (monitored daily using a nitrate-selective electrode). Seventeen seedlings per species were grown during 21 days. Non-destructive relative growth rate was determined individually on a fresh mass basis: at day 7 after transplantation, seedlings were removed from the growing medium, and roots were carefully drained but not dried to ensure subsequent growth. Total individual fresh mass was then assessed and seedlings were put back in the growing medium. At day 21, seedlings were harvested and weighted to determine total fresh mass. RGR$_{\text{max}}$ was calculated as the slope of the regression of ln(fresh plant mass) on time. The validity of this fresh mass-based RGR$_{\text{max}}$ was verified in a classic destructive growth analysis set up in parallel on 10 other species, of which five were common with this study. The RGR$_{\text{max}}$ rankings did not change significantly between destructive (dry mass-based RGR$_{\text{max}}$) and non-destructive estimations (Spearman’s $r = 0·96$; $P < 0·001$; $n = 10$).

**DATA ANALYSIS**

Anovas were performed on logarithmically transformed data to test the main effects of successional status and botanical family on each trait. Multiple comparisons among pairs of means were made using the Student–Newman–Keuls studentized post hoc test. Bivariate correlations were evaluated using Spearman’s rank coefficients on untransformed trait values. In PIC analyses, one-tailed sign tests were performed; cumulative $P$ values were calculated according to Sokal & Rohlf (1995). Due to the small sample size, the significance threshold for this sign test may not be reached. We therefore report attribute shifts in each PIC as + for an increase or – for a decrease as succession proceeds, to detect a majority trend. Finally, a principal components analysis (PCA) was performed to characterize the multivariate pattern of correlations among variables.

All the analyses were carried out with SAS (SAS Institute v.8, Cary, NC, USA).

**Results**

**TRAIT VARIATION AMONG SPECIES**

For all traits, species from early and intermediate stages were statistically indistinguishable from one another (Table 2; Fig. 2). At the end of the incubation period (8 weeks) the %MR ranged from 74·2% for Brachypodium phoenicoides to 26·5% for Dipsacus fullonum. Fits to the single exponential decay model were all highly significant (Table 2). The ANOVA indicated significant differences in this exponential decay ($K_{\text{pot}}$)

### Table 2. Traits of the 12 species studied

<table>
<thead>
<tr>
<th>Successional stage/species</th>
<th>SLA$_{\text{act}}$ (m$^2$ kg$^{-1}$)</th>
<th>LDMC$_{\text{act}}$ (mg g$^{-1}$)</th>
<th>LNC (mg g$^{-1}$)</th>
<th>LCC (mg g$^{-1}$)</th>
<th>LPC (mg g$^{-1}$)</th>
<th>TPh (mg g$^{-1}$)</th>
<th>RGR$_{\text{max}}$ (g g$^{-1}$ day$^{-1}$)</th>
<th>$K_{\text{pot}}$ (g kg$^{-1}$ day$^{-1}$)</th>
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<tr>
<td><strong>Early (E)</strong></td>
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<tr>
<td><em>Avena barbata</em> (Ab)</td>
<td>16·9 (0·7)</td>
<td>273 (7)</td>
<td>15·5 (1·8)</td>
<td>389 (10)</td>
<td>2·08 (0·04)</td>
<td>51·1 (2·83)</td>
<td>0·26 (0·02)</td>
<td>15·55 (14·43*** )</td>
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<tr>
<td><em>Bromus madritensis</em> (Bm)</td>
<td>32·8 (0·0)</td>
<td>240 (3)</td>
<td>24·5 (1·3)</td>
<td>408 (20)</td>
<td>1·77 (0·01)</td>
<td>24·4</td>
<td>0·26 (0·03)</td>
<td>14·30 (7·83*** )</td>
</tr>
<tr>
<td><em>Conyza sumatrensis</em> (Cs)</td>
<td>20·1 (0·8)</td>
<td>203 (6)</td>
<td>18·5 (1·1)</td>
<td>447 (50)</td>
<td>3·13 (0·07)</td>
<td>62·0 (0·85)</td>
<td>0·33 (0·03)</td>
<td>23·50 (7·78*** )</td>
</tr>
<tr>
<td><em>Crepis foetida</em> (CT)</td>
<td>17·9 (0·5)</td>
<td>162 (5)</td>
<td>17·2 (0·8)</td>
<td>439 (3)</td>
<td>3·22 (0·07)</td>
<td>78·1 (0·70)</td>
<td>0·33 (0·02)</td>
<td>29·08 (7·90*** )</td>
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<td><strong>Intermediate (I)</strong></td>
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<tr>
<td><em>Dactylis glomerata</em> (Dg)</td>
<td>26·0 (0·7)</td>
<td>294 (9)</td>
<td>21·8 (2·4)</td>
<td>452 (11)</td>
<td>3·34 (0·14)</td>
<td>26·2 (1·17)</td>
<td>0·22 (0·07)</td>
<td>12·12 (8·56*** )</td>
</tr>
<tr>
<td><em>Dipsacus fullonum</em> (Df)</td>
<td>19·2 (1·0)</td>
<td>179 (5)</td>
<td>13·1 (1·6)</td>
<td>437 (9)</td>
<td>3·06 (0·41)</td>
<td>55·3 (6·04)</td>
<td>0·27 (0·03)</td>
<td>36·19 (7·93*** )</td>
</tr>
<tr>
<td><em>Picris hieracoides</em> (Ph)</td>
<td>19·6 (1·2)</td>
<td>163 (5)</td>
<td>23·9 (2·1)</td>
<td>432 (5)</td>
<td>3·4 (0·56)</td>
<td>89·8 (9·53)</td>
<td>0·29 (0·03)</td>
<td>28·38 (5·43*** )</td>
</tr>
<tr>
<td><em>Sanguisorba minor</em> (Sm)</td>
<td>15·9 (1·1)</td>
<td>349 (7)</td>
<td>19·1 (1·5)</td>
<td>458 (0·3)</td>
<td>2·44 (0·09)</td>
<td>80·9 (2·26)</td>
<td>0·26 (0·03)</td>
<td>22·10 (11·09*** )</td>
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<tr>
<td><strong>Advanced (A)</strong></td>
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<tr>
<td><em>Agrimonia eupatoria</em> (Ae)</td>
<td>13·1 (0·4)</td>
<td>297 (4)</td>
<td>14·6 (0·8)</td>
<td>425 (18)</td>
<td>1·89 (0·08)</td>
<td>109 (1·85)</td>
<td>0·19 (0·03)</td>
<td>7·89 (14·20*** )</td>
</tr>
<tr>
<td><em>Brachypodium phoenicoides</em> (Bp)</td>
<td>13·1 (0·9)</td>
<td>411 (10)</td>
<td>10·9 (0·5)</td>
<td>437 (4·0)</td>
<td>2·19 (0·33)</td>
<td>9·32</td>
<td>0·18 (0·01)</td>
<td>6·39 (15·14*** )</td>
</tr>
<tr>
<td><em>Bromus erectus</em> (Be)</td>
<td>14·8 (0·8)</td>
<td>382 (7)</td>
<td>9·0 (0·3)</td>
<td>421 (14)</td>
<td>1·8 (0·06)</td>
<td>24·6</td>
<td>0·17 (0·02)</td>
<td>12·17 (10·11*** )</td>
</tr>
<tr>
<td><em>Rubia peregrina</em> (Rp)</td>
<td>14·4 (1·0)</td>
<td>230 (13)</td>
<td>13·1 (0·6)</td>
<td>417 (5)</td>
<td>1·97 (0·18)</td>
<td>35·6 (1·19)</td>
<td>0</td>
<td>8·50 (6·28*** )</td>
</tr>
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</table>

SE in parentheses; $n = 10$ for SLA$_{\text{act}}$ and LDMC$_{\text{act}}$, $n = 4$ for LNC, LCC and phenols (except for B. phoenicoides, B. erectus and *B. madritensis*: $n = 1$); $n = 15$ for RGR$_{\text{max}}$.

For $K_{\text{pot}}$, t-test values and test significance in parentheses.

*, $P < 0.05$; *, $P < 0.05$; ***, $P < 0.001$. Abbreviations follow Table 1.
according to successional stage, with early and intermediate successional species decomposing more rapidly than species from advanced successional stages ($F_{2,9} = 8.45, P = 0.009$; Fig. 2a).

RGR$_{\text{max}}$ varied among species from 0.17 day$^{-1}$ (*Bromus erectus*) to 0.33 day$^{-1}$ (*Crepis foetida* and *Conyza sumatrensis*). Species from early and intermediate successional stages had higher RGR$_{\text{max}}$ than species from the late successional stage ($F_{2,8} = 16.38, P = 0.001$; Fig. 2b).

SLA$_{\text{sat}}$, LNC and LPC showed the same trend along succession: species from early and intermediate stages
had significantly higher SLA_{sat} (F_{2,9} = 4.62, P = 0.041), LNC (F_{2,9} = 5.43, P = 0.028) and LPC (F_{2,9} = 4.77, P = 0.039) than species from advanced stages (Fig. 2c,e,g).

The trend in LDMC_{sat} (F_{2,9} = 2.32, P = 0.153) was in the opposite direction during succession, but was not statistically significant (Fig. 2d). Neither LCC (F_{2,9} = 2.06, P = 0.183) nor total phenols of green leaves (F_{2,9} = 0.31, P = 0.741) showed any significant differences between successional stages (Fig. 2f,h). However, we found a significant family effect on total phenols, with species belonging to Rosaceae and Asteraceae having higher concentrations of total phenols in green leaves (F_{3,7} = 12.90, P = 0.005).

PIC analyses were in accordance with the above results. In four of five contrasts, species from earlier successional stages had leaves with higher SLA_{sat} and LNC and lower LDMC_{sat} than their PIC partners from advanced succession (Fig. 2c–e). For all contrasts, species from early and intermediate succession had higher RGR_{max}, and leaves of earlier successional species decomposed more quickly than leaves from species from more advanced succession stages. No pattern was detected for LCC, LPC and total phenols.

LDMC_{sat} was negatively correlated with both K_{pot} and RGR_{max} (Fig. 3a,b): the litter of fast-growing species, with low LDMC_{sat} tends to decompose more rapidly than that of slow-growing species with high LDMC_{sat}. As a consequence, K_{pot} and RGR_{max} were significantly correlated to one another (Fig. 4). No significant correlations were found between total phenols, litter decomposability and RGR_{max}, and K_{pot} was not correlated to any of the leaf chemical fractions (data not shown). LNC was significantly correlated with SLA_{sat} (r = 0.77, P < 0.01) but not with other traits. LPC was significantly correlated to LCC (r = 0.68, P < 0.05). Finally, RGR_{max} was positively correlated with LPC (r = 0.62, P = 0.05).

**MULTIVARIATE RELATIONSHIPS**

The first axis of the PCA accounted for nearly 48% of variation in the data, while the second axis accounted for 22% (Fig. 5a). Axis 1 was determined by four traits: LPC, RGR_{max} and K_{pot} opposed to LDMC_{sat}. The second axis was determined by SLA_{sat} and LNC. A supplementary part of variation of the second axis was provided by total phenols, but this variable was not significantly correlated with the other variables. The third axis (15% of variance explained) was determined by LCC.

Loadings of species on axis 1 varied significantly between species of different ecological status (F_{2,9} = 8.51, P < 0.05; Fig. 5b): early and intermediate successional species had low values of LDMC_{sat} and high LPC, and grew and decomposed more quickly than species originating from advanced successional stages. Species belonging to the Poaceae family are discriminated from those belonging to other families. PIC tendencies confirmed the trait covariation according to species successional status.

**Discussion**
traits measured, but these two stages were generally distinct from the advanced stage. A possible explanation for this result is that the difference in age since abandonment for these two stages (3 vs 10 years) is smaller than between the intermediate and advanced stages (10 vs 25 years). This is an interesting result, which shows that functional stability is conserved in the first two stages despite a large change in floristic composition, while the progression towards the more advanced stage shows both specific and functional shifts. In the following discussion, the early and intermediate successional stages are referred to collectively as ‘early successional stage’ to account for this functional stability.

Complex litters produced by early successional communities were shown to decompose more rapidly than those produced by communities from more advanced stages (Garnier et al. 2004). A major finding of the present study is that this trend is also found for the most abundant species that compose these communities: species from early stages tend to decompose more rapidly than species from more advanced stages (Fig. 2a).

The decrease in \( K_{pot} \) with species’ successional status goes with a decrease in \( R_{GR_{max}} \) (Fig. 2). This is consistent with Tilman’s (1990) hypotheses on the role of the underlying environmental constraints of successional habitats (access to disturbed sites, availability of limiting soil resources, availability of light, etc.), which should lead to trade-offs in maximal growth rates of the species. A high relative growth is a prominent trait characterizing the ‘ruderal syndrome’ associated with disturbed environments (Grime 1979) and early stages of succession (Gleeson & Tilman 1994). Such a high rate is assumed to be adaptive in this environment, where rapid growth is both possible because of low competitive interactions with an established vegetation, and necessary to produce numerous seeds rapidly, leading to a high colonization potential. \( R_{GR_{max}} \) usually declines along successional gradients (Gleeson & Tilman 1994) as the frequency of disturbance decreases and/or the absolute or relative levels of resources change. Such changes in potential growth are mirrored in the field by changes in traits related to resource capture, such as SLA and LNC (cf. Wright et al. 2004); as found here (Fig. 2), these two traits tend to decrease as succession proceeds (Reich et al. 1995; Llambi et al. 2003). Our results also show lower LPC in species from advanced stages (Fig. 2), but we do not know whether this trend is general (cf. Reich et al. 1995). The two other leaf traits measured (LCC and phenols) did not show any trend with successional status.

Overall, our findings suggest that \( K_{pot} \) can be considered part of the fast–slow growing syndrome, although previous tests have produced mixed results (Cornelissen & Thompson 1997 for herbaceous species; Cornelissen et al. 1998 for trees).

Although our species selection was based on abundance in the successional sere rather than on taxonomy (non-random taxon sampling sensu Ackerly 2000), we were able to construct PICs for a subset of the species studied (Fig. 1). There were clear differences in trait values among PICs (Fig. 5b), but most conclusions on successional trends did not depend on the species pool taken for comparison (Fig. 2); we are therefore confident that these trends correspond to actual shifts related to the ecology of species, and not to the replacement of a particular taxonomic group by another one as succession proceeds.

FUNCTIONAL RELATIONSHIPS AMONG TRAITS

In our study, \( LDMC_{sat} \) of green leaves was strongly correlated with \( R_{GR_{max}} \) and \( K_{pot} \). LDMC relates to leaf
density (Shipley & Vu 2002; Niinemets & Kull 2003) and anatomy (Garnier & Laurent 1994). More precisely, LDMC_{sat} reflects the amount of mesophyll vs structural compounds (cf. Garnier & Laurent 1994; Van Arendonk & Poorter 1994) in the leaf. A high LDMC_{sat} corresponds to a low proportion of mesophyll and epidermis (light tissues) and a high proportion of vascular tissues and sclerenchyma (dense tissues) (Dijkstra & Lambers 1989; Niemann et al. 1992; Garnier & Laurent 1994). In terms of chemical composition, this corresponds to leaves rich in (hemi)cellulose, insoluble sugars and lignin (Poorter & Bergkotte 1992).

A high investment in assimilatory tissue (mesophyll) is expected to lead to a high photosynthetic and growth potential, in relation to a high concentration of nitrogen-rich, photosynthetically active mesophyll protoplast (Wright et al. 2004). Negative relationships have been found between LDMC_{sat} and/or tissue density and RGR_{max} in a number of studies (Garnier & Laurent 1994; Van Arendonk & Poorter 1994). A high LDMC_{sat} corresponds to leaves rich in (hemi)cellulose, insoluble sugars and lignin (Poorter & Bergkotte 1992).

The strong correlation between LDMC_{sat} and K_{pot} suggests that, as argued previously (Grime & Anderson 1986), structural traits of living leaves persist in litter. In order to understand the links between the properties of green leaves and initial litter, initial litter labile compounds (labile compounds = organic matter = (hemicellulose + cellulose + lignin)) of the 12 species were estimated using near-infrared spectroscopy (Gillon et al. 1999). Litter labile compounds were significantly correlated with LDMC_{sat}, RGR_{max} and litter decomposability (data not shown). Species that have a high production of biomass (high RGR_{max}, low LDMC_{sat}), produce litter rich in labile compounds and thus decompose more quickly. Leaf total phenols were not correlated either with RGR_{max} or with decomposition rate, in contrast with other studies (Almeida-Cortez et al. 1999). Generally, plant polyphenols affect decay rate as they can directly influence groups of soil organisms and prevent the breakdown of litter (Hättenschwiler & Vitousek 2000). A strong control on decay rate by phenolics-related compounds was found by Aerts & de Caluwe (1997). The lack of a relationship between litter decay rate and leaf total phenols in our study is probably due to the fact that our analyses were not very specific. As a consequence, the compounds analysed might correlate poorly with actual polyphenolics, which constitute the relevant fraction involved in the decomposition process (Schultz 1988).

These findings confirm the importance of some physical attributes of leaves (Cornelissen & Thompson 1997; Cornelissen et al. 1999) and litter (Gallardo & Merino 1993; Gillon et al. 1994) in decomposition processes. Our study further shows that these physical properties of leaves underlie the relationship between fast growth and fast decomposition rates. These properties appear to be captured adequately by LDMC_{sat}, a quantitative and easily measurable plant trait (Hodgson et al. 1999; Weiher et al. 1999), the ecological importance of which has been demonstrated in a number of studies (Wilson et al. 1999; Vendramini et al. 2002; Garnier et al. 2004). LDMC_{sat} can therefore be retained as a powerful functional marker (sensu Garnier et al. 2004) of litter decomposability and plant growth.

SCALING-UP TO ECOSYSTEM FUNCTIONING

In the study by Garnier et al. (2004), the decomposition rate of complex litters produced by multispecies communities was shown to decrease with field age, and the results from the present study show that trends are similar at the species level. To test how well field decomposition can be predicted with information derived from species, we combined the K_{pot} values obtained for the 12 species studied here with abundance data from the field plots studied by Garnier et al. (2004), and calculated an aggregated K_{pot-agg} value for each community as follows:

\[ K_{pot-agg} = \sum_{i=1}^{n} p_i \times K_{pot} \]

where K_{pot,i} is the K_{pot} of species i, p_i is the proportion of species i in the community, and n is the number of species taken into account in the calculation. The relationship between K_{pot-agg} and litter decay rate measured in the field (taken from Garnier et al. 2004) is positive and almost significant (r = 0.60, F = 0.068, n = 10), although for some fields the number of species for which K_{pot} is available is low compared with the total number of species present.

First, this suggests that mixing effects in litter produced by complex plant communities are small in comparison with interspecific differences measured for the species of this successional sere (Wardle et al. 2002; Wardle et al. 2003). Second, this agrees with the hypothesis stating that ecosystem functioning depends strongly on the attributes of species that compose these ecosystems (Chapin et al. 2000; Diaz & Cabido 2001; Lavorel & Garnier 2002), as applied to the specific case of litter decomposition.

Further tests are obviously required on a broader range of communities to check whether this finding is general.

Conclusions

For most traits measured, there were significant differences between the first two earliest successional stages and the more advanced one. Species from early successional stages grew quickly, producing leaves rich in nutrients with low dry matter content, and leaf litter that decomposed quickly. Later during succession, species had an opposite set of traits. This syndrome of traits was captured adequately by leaf dry matter content, which can therefore be considered as a powerful functional marker of species' functioning.
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