Functional Ecology

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New insights from multidimensional trait space responses to competition in two clonal plant species

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Running headline: Multidimensional trait space response to competition

Summary

1. Trait intraspecific variability determines community dynamics and species coexistence. In response to competition, plants can display intraspecific variability to enhance their competitive ability or stabilize their niche differences with competitors. This response is multidimensional because it involves changes along different functional axes and inevitable trade-offs between traits. Here, we transposed the recent concept of the multidimensional trait space to the analysis of intraspecific plant response to competition. We specifically tested the following: (1) in the absence of competitors, the plant multidimensional trait space will be packed towards strategies promoting plant colonisation, and (2) with competitors, the plant multidimensional trait space will be directed towards competition with its size and shaping characteristics dependent on competitor species richness.

2. We studied trait intraspecific variability of two clonal species, *Brachypodium pinnatum* (L.) P. Beauv. and *Elytrigia repens* (L.) Gould, in response to competition. We analysed plant response in the absence of competitors and in competition. Competition treatments included intraspecific and interspecific experimental mixtures with increasing species richness. For each target species and each treatment, we built an hypervolume based on six traits involved in the three-dimensional competition (i.e. ramet and connection traits). We measured these hypervolumes for their size, similarity and the contribution of traits in their shaping.

3. In the absence of competitors and for both species, we demonstrated a multidimensional trait space packing towards a colonisation strategy. Under competition, the multidimensional trait spaces of the two target species were the widest at the extremes of the richness gradient, *i.e.*, intraspecific and interspecific high richness competition treatments. High intraspecific variability either promoted niche differentiation from individuals of similar species or reflected the large range of competitive responses deployed when plants were faced with many different competitor identities. The multidimensional response process was based on

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fine adjustments of various traits depending on the surrounding neighbourhood composition and more specifically, on the competitor functional similarity with the target species.

4. This study emphasises the multidimensionality of species competitive response, and also underlines the so far neglected importance of competitor species richness for trait intraspecific variability and subsequently community assembly.

Introduction

Recent studies from trait-based ecology show that trait variability within plant species is of particular importance in determining community dynamics and species coexistence (Lichstein, Dushoff, Levin, & Pacala 2007; Whitlock, Grime, Booth, & Burke 2007; Jung, Violle, Mondy, Hoffmann, & Muller 2010; Messier, McGill, & Lechowicz 2010; Bolnick et al., 2011). Among various mechanisms responsible for this intraspecific trait variability (e.g. neutral processes, mutation, genetic local adaptation), phenotypic plasticity is the one supporting species ability to overcome abiotic and biotic filters such as competition, subsequently affecting community assembly processes (Jung et al., 2010).

The limiting similarity theory (MacArthur & Levins, 1967) predicts that individuals can only coexist if they use resources differently *i.e.* display distinct functional trait (Kraft, Godoy, & Levine 2015). Competitive plastic responses in plants relies then on a combination of traits as they compete for multiple resource, both above and belowground (Grime et al., 1997; Westoby, Falster, Moles, Vesk, & Wright 2002; Reich et al., 2003; Wright et al., 2007), leading to inevitable trade-offs and interactions between traits (Grime et al., 1997). As

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a result, a conceptual framework that based community ecology on multiple traits was recently proposed (Blonder, Lamanna, Violle, & Enquist 2014; Laughlin, 2014). By analogy to the concept of ecological niche (Hutchinson, 1957), this framework suggests to infer community assembly processes using the multidimensional space of species traits rather than single traits (Bonser, 2006). Kraft et al. (2015) demonstrated that for annual plants, multidimensional trait space allows a better understanding of the drivers of competition outcomes. Here, we thus suggest transposing this promising framework at the intraspecific level, to study perennial plant communities that include clonal plants. These plants are characterised by a plastic modular structure with vertically growing shoots (ramets) arranged in a branched network allowing the horizontal spread of the clone (Harper, 1981; Hutchings, 1999). In perennial plant communities, competition is then three dimensional as it occurs in the horizontal and vertical directions following clonal plant growth. Competitive responses would then involve complex modifications of aerial traits linked with resource use (vertical traits) but also of clonal traits involved in space acquiring (horizontal traits). These competitive responses may additionally be modulated by the clonal growth strategy of the individual (space colonisation *vs*. space pre-emption), as it determines the individual neighbourhood characteristics (Benot, Bittebiere, Ernoult, Clément, & Mony 2013).

Plant intraspecific response to competition may be determined at first by the single presence of competitors. In the absence of competitors, clonal plant growth is directed toward space acquiring and limiting intra-individual competition (Bittebiere, Garbey, Smaoui-Feki, Clément, & Mony 2014). This promotes strategies to improve space exploration, notably in the horizontal dimension through internode elongation (Bittebiere et al., 2014). The multidimensional trait space should then be packed toward a colonisation strategy based on traits mostly involved in the horizontal growth. In response to competitors' presence however, Novoplansky (2009) described three strategies (avoidance, confrontation, and

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tolerance), all possibly involving vertical and horizontal traits of clonal plants. Under competition, the multidimensional trait space should then converged toward one of these strategies or include all of them depending on the local competitive neighbourhood characteristics (Miller, Bowman, & Suding 2007; Violle et al., 2009; Ashton, Miller, Bowman, & Suding 2010; Bittebiere & Mony, 2015).

Numerous studies demonstrated that trait plastic responses (Turkington, Hamilton, & Gliddon 1991; Herben & Novoplansky, 2010; Bittebiere, Renaud, Clément, & Mony 2012), related to processes of stabilizing niche differences or increasing competitive ability (Chesson, 2000; Turcotte & Levine, 2016), depend on competitor identity. In particular for intraspecific competition, a niche differentiation process should limit the competition intensity through horizontal traits plasticity, allowing complementary ramet positioning (Herben, Krahulec, Hadincová & Pecháčková, 1994). However, only a few studies have investigated competitive responses in more complex multispecific assemblages (*i.e.*, more similar to real situations) (but see Bittebiere & Mony, 2015). In such multispecific assemblages, species richness modulates the probability of locally meeting species of varying identities and may then determine the plant response (*i.e.* variations in its multidimensional trait space). Under low species richness, competitive response of target plant should converge toward the most adaptive strategy in the presence of these few particular species. Multidimensional trait space will then be of small size with only a few traits driving the competitive response. Under high species richness however, target individuals may face a large range of local neighbourhood compositions, leading to varying responses of the target plant supporting its competitive ability and subsequently increasing the size of its multidimensional trait space. These responses must rely on both vertical and horizontal traits, because the displayed responses specifically depend on each competitive interaction.

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Based on this framework of the multidimensional trait space, we tested the effects of species richness (*i.e.* the number of competitor species) on the intraspecific variability of multiple traits. Species richness was manipulated in a large experimental design to study individual plant responses across a no competition treatment and one intra- and three interspecific competition treatments. We assessed the individual responses of two *Poaceae* species with contrasted clonal growth strategies (space colonisation *vs.* space pre-emption) and that were sampled in these five treatments. The intraspecific variability of individual responses was assessed through hypervolumes (*i.e.*, multidimensional trait spaces) built for each species using measurements of vertical and horizontal traits involved in the competitive response. We measured three indicators to compare the hypervolumes between competition treatments: i. hypervolume size, ii. hypervolume overlap calculated between pairs of treatments and indicating hypervolumes similarity, and iii. each individual trait contribution to the hypervolume shaping. Specifically, we used this experimental design to test the following hypotheses:

(1) In the absence of competition, the multidimensional trait space should be packed towards the strategies that promote plant colonisation. Hypervolume should then be narrow with low similarity with hypervolumes calculated for target plants in competition. The most contributing traits to hypervolume shaping are expected to be those involved in space colonisation (internode length, number of ramets and branches) to forage for free patches and prevent intra-individual competition.

(2) Under competition, the multidimensional trait space should reflect plant adjustments to competition but its size would vary with the type of competition (intra- vs. interspecific competition) and the species richness (Fig. 1). In addition, the number of traits involved in shaping the multidimensional trait space should increase with species richness.

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Materials and methods

Target species

Elytrigia repens (L.) Desv. and *Brachypodium pinnatum* (L.) Beauv. are two common perennial species in the grasslands of western Europe. Both species grow laterally by producing sympodial plagiotropic rhizomes of which the apexes develop into erected shoots (ramets) (CLO-PLA database, Klimešová & de Bello, 2009). Both species present intraspecific variation in their clonal and aerial traits, particularly in response to resource availability and competition (de Kroon & Knops, 1990; van der Werf, Visser, Schieving, & Lambers 1993; Pottier & Evette, 2009; Bittebiere & Mony, 2015).

Competition and no-competition treatments

To determine whether the intraspecific variability and therefore, the multidimensional trait space of the two target species changes across treatments, we cultivated on a homogenised mixture of sand (20%) and soil (80%), four competitive mixtures planted in May 2009, and 10 ramets of each species alone (referred to as the no-competition treatment). These five treatments were settled within the experimental garden of the University of Rennes 1 (France).

The richness of the four competitive mixtures varied from one to 12 competitor species, which corresponds to the range of species richness recorded in mesophilous grassland ecosystems in France (see e.g. Marion, Bonis, & Bouzillé 2010). This gradient allowed us to manipulate the probabilities of meeting each competitor species (Table S1). Specifically, the competitive mixtures were the following: (1) each target species in monoculture (only intraspecific competition); (2) each target species mixed with three other species, either *E. repens* with *Holcus mollis* L., *Agrostis stolonifera* L., and *Ranunculus repens* L. or *B. pinnatum* with *Festuca rubra* L., *Agrostis tenuis* Sibth., and *Anthemis nobilis*

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L. (*i.e.,* four competitor species); (3) a mixture of all eight of these species; and (4) a mixture composed of the previous eight species with four additional species (*Lolium perenne* L., *Dactylis glomerata* L., *Holcus lanatus* L., and *Centaurea nigra* L.) for a total of 12 competitor species (see Benot et al., 2013 and Bittebiere & Mony, 2015 for more details on the design). These species were selected from a set of species that are widely distributed in temperate grasslands in western France (des Abbayes, Claustres, Corillion, & Dupont 1971).

For each target species, the four competitive mixtures were replicated eight times, while the no-competition treatment was replicated 10 times. The replicates were randomly positioned within the outdoor experimental design. For the no-competition treatments, *E. repens* and *B. pinnatum* were each cultivated in 10 large pots. In competition, species were grown in 48 mesocosms (mixtures of eight and 12 species were shared between the two target species) that were $1.30 \times 1.30 \times 0.25$ m. Pots and mesocosm displayed similar heights to avoid constraints on the rooting depth. In each mesocosm, 48 ramets were initially transplanted and placed 16 cm apart, following a hexagonal pattern to ensure balanced competitive interactions (Birch, Oom, & Beecham 2007). All ramets were equally distributed over all the co-occurring species in the mixture, and the design then corresponded to replacement series, with the overall density of individuals within the mixture kept constant. The species positions in the plantation pattern were randomised for each mesocosm. Nevertheless, species covers changed over time due to the ongoing dynamics of the competitive mixtures and the clonal propagation of their initially planted ramets. The transplanted ramets of each species (including for *E. repens* and *B. pinnatum*) were collected from several locations in temperate grasslands near Rennes (Brittany, West France) to maximise genetic diversity among clones.

Each year, all the mature inflorescences were cut to suppress seed production, and all of the seedlings that emerged from the substrate seed bank were removed manually. The

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aboveground biomass was mown in late September. To prevent any possible effects of drought, the mesocosms and pots were watered every two days during the dry season (July–August).

Trait measurements

In April 2012 (i.e., after three years of competitive mixtures cultivation), we sampled each target species from five points within all mesocosms (in the centre and the four mesocosm corners when target species were sufficiently abundant to make this selection process possible). In total, we sampled 40 clonal fragments of each target species in each competitive mixture, and 10 clonal fragments from the no-competition treatment. The sampled fragments resulted from the clonal propagation of the initially planted *E.repens* and *B. pinnatum* ramets. They always began with one mature shoot to standardise sample age and were composed of five consecutive rhizome internodes and the associated shoots. We measured two types of plant traits. First, we measured vertical traits: ramet height, Specific Leaf Area (SLA), and Leaf Dry Matter Content (LDMC). These traits characterise plant photosynthetic efficiency and tissue conservation, *i.e.*, resource use (Cornelissen, Lavorel, & Garnier 2003). These traits were measured on the older mature ramet of the sampled fragment to standardise the age and position of the ramet along the rhizome, following Cornelissen, Lavorel, and Garnier (2003). Second, we measured horizontal traits: maximum internode length, the number of branches (*i.e.*, the number of rhizomes that branched from the sampled fragment rhizome), and the number of ramets (*i.e.*, potential new clonal individuals). These traits characterise space colonisation in clonal species.

Constructing six-dimensional hypervolume

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We built the six-dimensional trait space of *E. repens* and *B. pinnatum* using a multidimensional kernel density estimation procedure (Blonder et al., 2014). This method allows a hypervolume to be constructed with an arbitrary number of dimensions (*sensu*, number of traits), which in this case, represented the multidimensional trait space of the target species. We constructed one hypervolume for each species in each treatment (*i.e.*, the four competition treatments and the one no-competition treatment) using a kernel based on the Silverman estimator (Silverman, 1992). Before analysis, we checked for redundancy between traits (no correlation between traits with coefficients higher than 0.6, fig. S2), and all trait data were centred and scaled (*sensu*, data were standardised using the mean and standard deviation of the data from all treatments simultaneously, Blonder et al., 2014). All hypervolumes were constructed using the "hypervolume" package from R 3.2.4. (Blonder et al., 2014).

Effects of treatment on the multidimensional trait space of target species

To determine treatment effects on target species hypervolumes, we first calculated their volumes in the four competition treatments and in the no-competition treatment. Volume is a measure of hypervolume size and represents the width of the multidimensional trait space of the species (*sensu*, the variability of all traits shaping the hypervolume simultaneously). Because we could only calculate one hypervolume per target species and treatment (hypervolume calculation requires at least one individual per each trait involved in hypervolume shaping), we lacked variability for comparing volumes between treatments. To solve this problem, we simulated 1000 hypervolumes per target species and treatment using bootstrapping (Gardener, 2014). Specifically, we made a random selection of sampled fragments with replacement. Because the volume depends on the number of values used to build the hypervolume (Lammana et al., 2014), we set the number of values used to build the

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hypervolume at 10 for each simulation, which was the number of values obtained in the treatment with the least sampled fragments (*i.e.*, no-competition). Therefore, we compared the volume of different treatments with all the hypervolumes built from the same number of values. For each competition treatment, because we used 10 ramets sampled from both similar and different mesocosms for hypervolume calculation, hypervolume simultaneously included trait variation within and among mesocosms. Thus, to evaluate the relative importance of intra-treatment and inter-treatment effects, we compared the variability of the hypervolumes built for each treatment and target species against the variability of a hypervolume built using ramets from all treatments at the same time. As the variability of intra-treatment hypervolumes was always negligible compared with inter-treatment variability (Fig. S3), we assumed that the observed hypervolume variations were actually due to effects of competition treatments.

Second, we calculated the overlap between the hypervolumes of target species for each pair of treatments (*i.e.*, the four competition treatments and the no-competition treatment). The overlap between two hypervolumes measured the intersection of the hypervolumes in the six-dimensional volume defined by the traits and was calculated as multidimensional variation of Sorensen similarity index:

$$Overlap = 2 \times \frac{volume\ shared\ by\ hypervolume\ A\ and\ B}{volume\ of\ hypervolume\ A + B}$$

In this case, hypervolume overlap represented the similarity of the two multidimensional trait spaces (*sensu*, the overall similarity of all trait values between the two hypervolumes), with high overlap indicating more similar spaces. Similarities between multidimensional trait spaces were then compared using a Ward's clustering method (Murtagh & Legendre, 2014). We also calculated overlap values between the hypervolumes of all species from the competitive mixtures to check that the target and all competitor species occupy different multidimensional trait spaces (Table S2).

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Effects of treatments on traits shaping the hypervolumes of target species

We calculated in each of the five treatments the contribution of the six measured traits to multidimensional trait space. The trait contribution is related to its variability, relatively to other traits. An increase in trait variability leads to an increased axis range in the hypervolume. Thus the more variable is one trait relative to the five others, the more it contributes to the hypervolume shaping *i.e.* is more important for the multidimensional trait space definition. Trait contribution is calculated as the ratio between the volumes of two hypervolumes: the hypervolume which considers all the traits, and the hypervolume which considers all the traits but the trait of interest. For each target species separately, we compared the measured contribution of each single trait in shaping the hypervolumes under the different treatments to an expected value. Specifically, we simulated 1000 hypervolumes for each species by resampling 40 random clonal fragments from the pool of all the treatments (similar to that presented in Lammana et al., 2014). These simulated hypervolumes represented the potential multidimensional trait spaces of species in our experiment, because they were created from the trait values extracted from all of the observed values. Then, for each treatment, we compared the contribution of each trait against a 95% confidence interval built from the simulated data (*i.e.*, by selecting the 2.5 and 97.5 percentiles of simulated values). Thus, when the value of contribution of any trait was outside the confidence interval, the contribution of that trait in that treatment was significantly different from that expected for the species in the experiment.

Results

Influence of treatments on target species multidimensional trait space

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To determine the effect of competition on species hypervolumes (*i.e.*, their multidimensional trait spaces), we compared the sizes (volumes) and similarity (overlap values) among the five treatments (four competition treatments and one no-competition treatment). Our results suggest that the volumes of *E. repens* and *B. pinnatum* hypervolumes varied among treatments (Fig. 2). The smallest hypervolumes of target species were in the absence of competitors compared to the four competition treatments.

Under competition, target species consistently tend to display larger hypervolumes in treatments with one and 12 competitor species than those in the four and eight species treatments. Overlap analyses were similar for both target species and showed that the largest dissimilarity in hypervolume was between the no-competition treatment and the four competition treatments (Fig. 3). Two pairs of competition treatments were more similar than others: those with one and 12 competitor species vs. with four and eight competitor species.

Influence of treatments on main traits shaping hypervolumes

In the absence of competitors, for both target species *E. repens* and *B. pinnatum*, we found that two of the three traits involved in space colonisation (*i.e.*, number of ramets and branches) contributed significantly more to the hypervolume shaping than expected (Table 1). Conversely, traits involved in resource use were overall less contributing than expected.

Under competition and for both target species, only one trait (number of ramets) did not display a response in any competition treatment (Fig. 4).

In the intraspecific competition treatment, the response depended on the target species. In *B. pinnatum*, no trait had higher, and only SLA had lower contribution to hypervolume shaping than expected (Fig. 4b). By contrast, in *E. repens*, ramet height and number of branches on the one hand, and maximum internode length on the other hand were

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respectively significantly more and less contributing to hypervolume shaping than expected (Fig. 4a).

In interspecific competition, the number of branches was less contributing to hypervolume shaping than expected in almost all competition treatments for both *E. repens* and *B. pinnatum* (with the exception for *E. repens* in the 12 competitor species treatment) (Fig. 4). Except for the number of branches, patterns of trait contribution to hypervolume shaping strongly varied with the competition treatment and between target species with no apparent relationship with the gradient of species richness. In *E. repens*, no trait was more contributing than expected (Fig. 4a), whereas in *B. pinnatum*, either LDMC or ramet height and SLA were more contributing than expected in the four and the 12 competitor species treatments, respectively.

Discussion

The multidimensional trait space primarily depends on the presence of competitors

In both target species, the primary determinant of multidimensional trait space was the single presence of competitors. As expected, the multidimensional trait space tends to be packed in the absence of competition. This space was strongly dissimilar from the others, indicating a shift in plant strategy from space colonisation to varying competitive strategies. In absence of competitors, individuals mostly seek colonizing space, thereby generating environmental heterogeneity related to ramet spatial distribution and density (Jackson & Caldwell, 1993; Herben, During, & Law 2000). The most important traits for shaping the multidimensional trait space were then the number of branches and ramets. In heterogeneous space, the high flexibility in the number of branches helps to reach free spaces (Slade & Hutchings, 1987; Sutherland & Stillman, 1988) and avoid intra-clonal competition (Lovett-Doust, 1981b; Schmid, 1986), whereas changes in the number of ramets indicate different degrees of spatial

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aggregation depending on free-patch size. By contrast, values of the traits involved in resource use converged. Individual plants constantly displayed low SLA and ramet height or LDMC, suggesting they invested less in light acquiring organs variability (Figs. S4, S5). Optimal ramet positioning through horizontal trait plasticity may cancel the requirement of a foraging response in the vertical direction.

The multidimensional trait space under competition depends on the number of competitor species in the local neighbourhood

The multidimensional responses of both *E. repens* and *B. pinnatum* depended on the competition treatment and followed a similar pattern. The largest multidimensional trait spaces were in the intraspecific and 12 species treatments, whereas these spaces were intermediate in the four and eight species treatments. Additionally, hypervolumes were more similar between the four and eight species treatments and between the intraspecific and 12-species treatments.

Under intra- and interspecific competition, trait variability is likely related to mechanisms of equalizing fitness and stabilizing niches differences (Chesson, 2000). Under intraspecific competition, the target individual and its neighbours have similar resource use. Strong plasticity would thus result from a niche differentiation process following the limiting similarity prediction (MacArthur & Levins, 1967; Violle et al., 2012). Plastic individuals may then differ from their functionally similar neighbours to limit competition intensity and support coexistence. Under interspecific competition, trait variability increases while competitive responses change, from the four and eight species to the richest (12 species) community. This species richness effect was likely due to the probabilities of encountering and then locally interacting with competitors of different identities (i.e., resource acquisition strategies) and competitive abilities (Table S1). Accordingly, target species should then adopt

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and combine multiple strategies [avoidance, confrontation, tolerance, *sensu* Novoplansky (2009)] to adjust to these varying competitor identities (Bittebiere et al., 2012; Callaway, Pennings, & Richards 2003) and support their fitness differences (Turcotte & Levine, 2016).

Our results thus underline the strong role of intra- vs. interspecific competition, and for the first time, highlight the probable role of species richness for trait intraspecific variability in the plant community. This was so far little considered, as experimental community ecology is mostly based on simplified plant species assemblages. Ultimately, considering species richness in future works dealing with plant community assembly rules should constitute a huge step forward in determining mechanisms supporting or hindering species coexistence.

Trait importance in the shaping of multidimensional trait spaces along the gradient of species richness

In contrast to our expectations, the number of traits involved in the target species multidimensional response to competition was not influenced by the gradient of species richness.

Under intraspecific competition, the niche differentiation process not only relies on horizontal traits. The characteristics of the multidimensional trait space were partially shared with the no-competition treatment. Notably, the number of branches remained highly variable relatively to other traits, in *E. repens*, unlike in *B. pinnatum*. Monocultures of guerilla individuals must, at a very local scale (a few centimetres), be similar to the no-competition treatment because of the rapid and diffuse clonal growth of guerilla species. However, the clumped clonal growth of *B. pinnatum* [*i.e.* phalanx species, (Lovett Doust, 1981; Benot et al., 2013)] generated more heterogeneous and far less predictable competitive environments in monocultures (Bittebiere et al., 2012) than in the no-competition treatment. Thus,

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individuals did not invest in the variability of one particular trait (Alpert & Simms, 2002). Instead, the response of *B. pinnatum* in intraspecific competition relied on the local adjustment of different traits.

In interspecific competition, the number of branches converged to low values in both target species (Figs. S4, S5). In these growing conditions, competitive ability is indeed supported by a switch of plant growth toward a caespitose form (consolidated plant positions). Nevertheless, with the exception of the number of branches, the two target species displayed different multidimensional responses to interspecific competition. In *E. repens*, no particular trait was more plastic than expected. Individual responses to the local neighbourhood involved different traits, to specifically adjust to each local competitor identity, resulting in a wide multidimensional trait space but with no particular trait characteristics. By contrast, in *B. pinnatum*, we overall found (with four and 12 competitor species) that traits involved in resource use were the most plastic. Because of its phalanx growth strategy, *B. pinnatum* displays low dispersal ability (Benot et al., 2013), which highly conditions its neighbourhood composition. In our system, *B. pinnatum* individuals more often met competitors from the most abundant species (see Table S1, although abundances at the mesocosm scale are a proxy of the individual neighbourhood). Some of the most abundant species in the four and 12 competitor species treatments are functionally similar to *B. pinnatum*: *A. tenuis* shared close LDMC values, while *E. repens* and *D. glomerata* both displayed similar SLA and ramet height values (Table S3). Functional differentiation from these species might explain the importance of traits involved in resource use in shaping the multidimensional trait space of *B. pinnatum*. Besides, investment in the plasticity of *B. pinnatum* ramet height relatively to other traits, increases with competitor species richness, likely because of an asymmetric exploitation of light resource (DeMalach, Zaady, Weiner, & Kadmon 2016). This trait would support *B. pinnatum* fitness differences with neighbours.

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Manipulating species richness resulted in nonlinear variations of competitor meeting probabilities across competitive treatments (Table S1), which subsequently led to complex multidimensional responses in relationship with competitor relative abundances and functional similarity with the target species. Competitive responses under interspecific competition eventually relied on fine adjustments of multiple traits involved in either space exploration or resource exploitation depending on the local neighbourhood.

The determining role of trait intraspecific variability in competitive environments

This study is the first to determine the intraspecific variability of the multidimensional trait space notably in response to competition. We demonstrated that the competitive neighbourhood structured this space, mostly by the presence of competitors alone, and to a lesser extent by their species richness. The multidimensional trait space is determined by species richness likely because it influences the probability of meeting competitors of different identities. The amplitude of the overall trait variability followed a curved relationship with the highest values at both extremes of the gradient characterizing two main processes of competitive response: niche differentiation from functionally similar neighbours (variations in resource use) and specific plastic adjustments to the various competitor species supporting competitive abilities (variations in resource acquisition strategies). In our experimental design, we used the same competitor species in all the replicates at each level of our species richness gradient; thus, the effects of richness and competitor species identities on the multidimensional response to competition were partially confounded. Therefore, we recommend that further studies in community ecology based on species richness gradients use different species at similar levels of richness to solve this problem.

The shaping of the multidimensional trait space under intraspecific competition, *i.e.*, with functionally similar competitors, depended on the clonal growth strategy shared by the

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target plant and its neighbours. Under interspecific competition, the competitive response relied on fine adjustments of multiple traits involved in environmental exploration and exploitation depending on the local neighbourhood. These fine adjustments suggested that the competitive response was integrated at the scale of the individual. Overall, highlighted by the results, we argue that to assess with accuracy the evolutionary and ecological processes that determine plant resource acquisition strategies and support species coexistence within communities, a large range of traits and their fine variations must be considered.

This work demonstrates the importance of trait intraspecific variability in plant responses in a competitive environment. Beyond contributing to the current debate on the relative importance of intra- *versus* interspecific trait variation in plant community assembly, we provide new insights on the underlying mechanisms generating this variability. Our results suggest that trait intraspecific variability is selected for a particular target plant species relative to the composition (species richness) of its plant neighbourhood. Nevertheless, its consequences for the whole plant community dynamics remain to be determined as phenotypic plasticity in response to competition can either hinder or favour species coexistence (Turcotte & Levine, 2016).

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Authors' contributions

AKB and CM conceived the ideas and designed the methodology; AKB collected the data; HS analysed the data; AKB, HS and CM wrote the manuscript.

Data availability

All data are available in figshare (DOI: 10.6084/m9.figshare.6860138; https://figshare.com/s/b0a0e77f4416af93776c).

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Table

Table 1. Trait contributions to hypervolume shaping for the target species in the no-

competition treatment.

Target species	Traits	Observed value	$Sim_{0.025}$	$Sim_{0.975}$	Result
E. repens	Maximum internode length	2.64	1.96	2.94	ns
	Number of branches	4.07	1.91	2.91	+
	Number of ramets	4.56	1.70	3.13	+
	Ramet height	0.96	1.90	2.98	-
	LDMC	2.79	1.66	3.19	ns
	SLA	1.08	1.88	2.89	-
B. pinnatum	Maximum internode length	2.69	1.84	2.99	ns
	Number of branches	2.87	2.12	2.85	+
	Number of ramets	3.34	1.82	2.99	+
	Ramet height	2.38	2.06	2.94	ns
	LDMC	1.35	1.77	2.91	-
	SLA	1.13	2.16	2.88	-

Notes: $Sim_{0.025}$, 2.5 percentile of the simulated data; $Sim_{0.975}$, 97.5 percentile of the simulated data. When comparing the observed value with the simulated data, trait contribution was significantly higher (+), lower (-), or not different (ns).

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Figures

a. Trait intraspecific variability : multidimensional space shift involving trade-offs between individual plant traits

Multidimensional trait space A **Multidimensional trait space B**
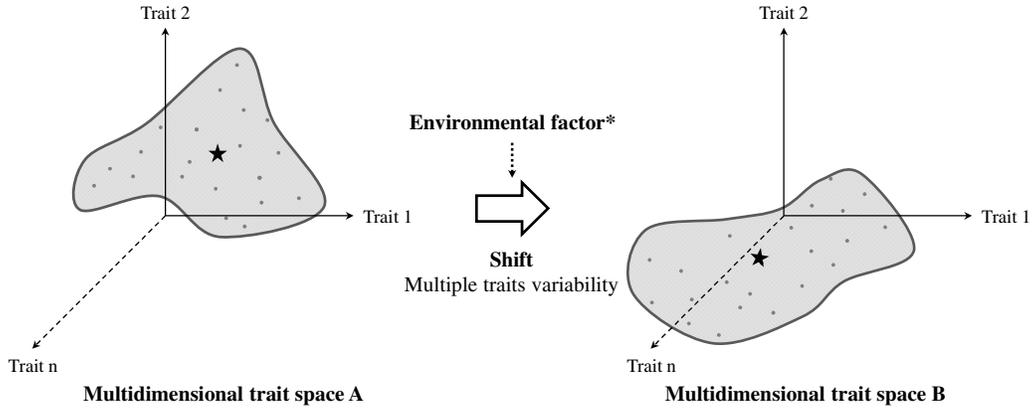
b. Response to competition along a gradient of community richness : multidimensional trait spaces of similar barycentres but varying volumes
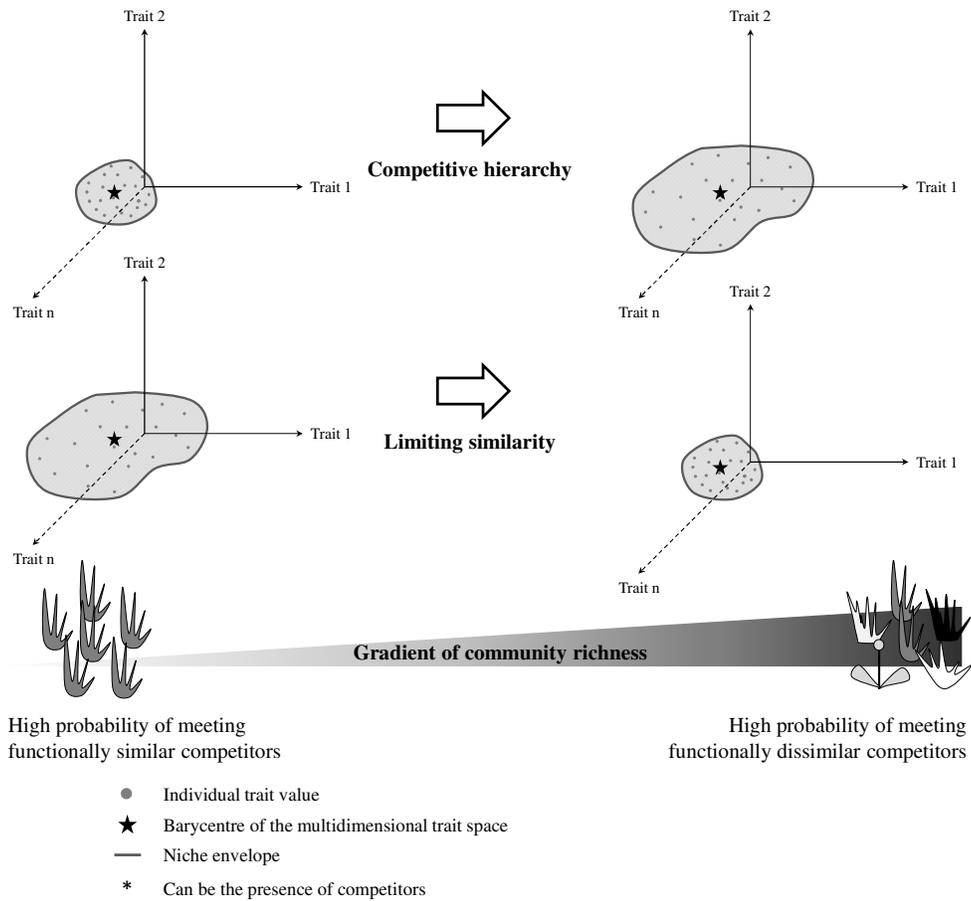

High probability of meeting functionally similar competitors

High probability of meeting functionally dissimilar competitors

- Individual trait value
★ Barycentre of the multidimensional trait space
— Niche envelope
* Can be the presence of competitors

Figure 1. Conceptual framework of the study. Theoretical effects of competition on the multidimensional trait space of species.
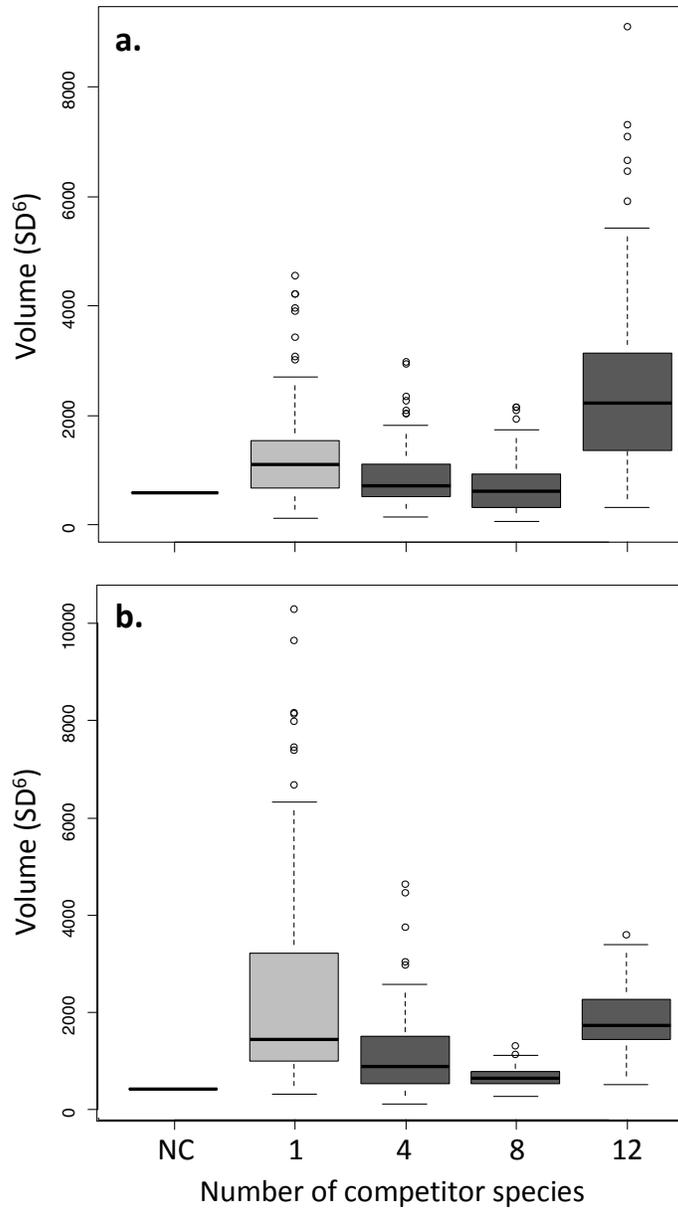
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Figure 2. Distributions of volumes of target species hypervolumes depending on treatment: (a) *Elytrigia repens*, (b) *Brachypodium pinnatum*. Volume indicates hypervolume size based on the six traits considered. NC, No-competition treatment (white boxplot); Competition treatments (light and dark boxplots): 1: *E. repens* or *B. pinnatum* in monoculture (intraspecific competition); 4, 8, and 12: *E. repens* or *B. pinnatum* in mixture with four, eight, and 12 competitor species (interspecific competition), respectively.
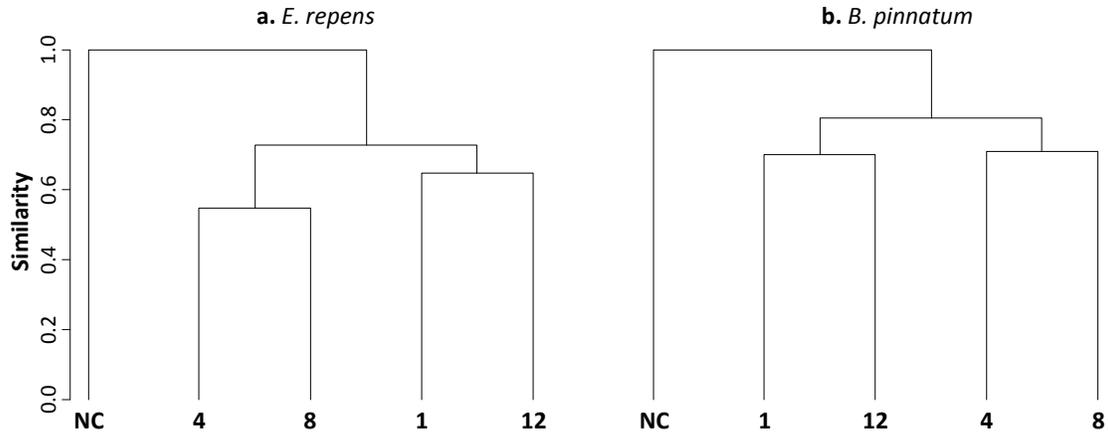
Figure 3. Similarity between the hypervolumes of the target species depending on treatment (no-competition and one, four, eight, and 12 competitor species): (a) *Elytrigia repens*; (b) *Brachypodium pinnatum*. The one competitor species treatment corresponds to intraspecific competition. The Y-axis represents 1-Overlap (overlap indicates similarity between hypervolumes). Smaller branch lengths represent treatments with similar hypervolumes. Dendrogram was created following Ward's clustering method (Murtagh & Legendre, 2014).
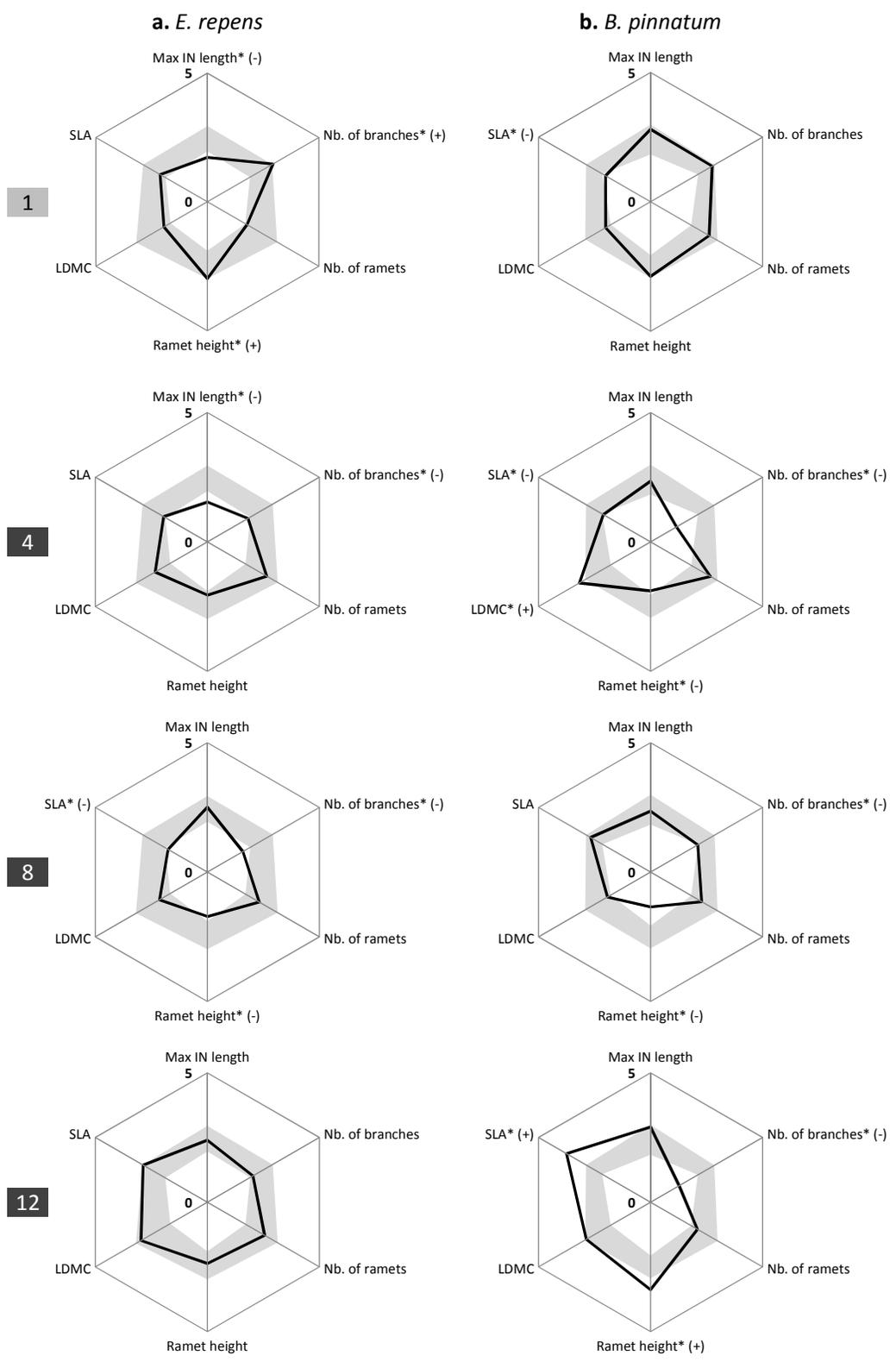
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a. *E. repens*   **b.** *B. pinnatum*

1

4

8

12

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Figure 4. Contribution of target species traits to hypervolume shaping, in the intra- (one competitor species) and interspecific competition treatment (four, eight, and 12 competitor species): (a) *Elytrigia repens*; (b) *Brachypodium pinnatum*. For information on the contributions of target species traits in the no-competition treatment, see Table 1. Trait contribution to hypervolume shaping is related to its variability relatively to the five other traits. The grey area represents the confidence interval of trait values built from the simulated target species hypervolume in the experiment by selecting the 2.5 and 97.5 percentiles of simulated values (see Materials and methods section for details). Stars indicate when the observed value of trait contribution is significantly higher (+) or lower (-) than expected through simulations. Max. IN length: maximum internode length; Nb. of branches: number of branches; Nb. of ramets: number of ramets.

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