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OIKOS

Research

Dissecting fine root diameter distribution at the community level captures root morphological diversity

Amandine Erktan, Catherine Roumet and François Munoz

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Characterizing plant functional diversity is essential to decipher community assembly rules and ecosystem functioning. Most studies focused on above-ground traits whereas the analysis of root diversity lags far behind. We analyzed the structure of fine root (< 2 mm) diameter distribution at the community level as an indicator of root morphological diversity, and hypothesized that the shape of the distribution provide insights on root types and root exploration strategies. We tested this hypothesis along a successional gradient (6–69 year-old) with yearly mowing to better understand assembly rules regarding to belowground processes, and explored the relations between the parameters describing its modes and the vegetation composition and ecological properties of plant communities. Most communities showed a multimodal distribution, with two main modes corresponding to absorptive roots (thinner root mode) and transport roots (coarser root mode), and a third mode of lower importance corresponding to large transport roots. In early succession, the thinnest root mode was prominent, reflecting the dominance of thin absorptive roots and a low proportion of transport roots, resulting in a low root morphological diversity. As succession proceeds, the relative proportion of the second mode increased, and the proportions of the two main modes were more balanced, resulting in an increased variance and root morphological diversity. Furthermore, the first root mode (absorptive roots) became wider and shifted from very thin to thicker roots, suggesting the coexistence of various root strategies for resource exploration. Yearly mowing did not affect root diameter distribution, which may relate to the relative low mowing pressure that enabled woody species to remain, with stunted stature. Overall, our study demonstrates that the distribution of fine root diameters sheds light on root morphological diversity at the community level, and provide hints on the co-existence of root types and strategies for resource use and exploration.

Keywords: diversity, ecological succession, fine root diameter distribution, root traits

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Introduction

Characterizing plant functional diversity is essential to understand community assembly rules (Bruelheide et al. 2018) and ecosystem functioning (Zirbel et al. 2017). Plant traits have been widely used to assess functional diversity at the community level (Garnier et al. 2004, Gross et al. 2007, Buzzard et al. 2016). However, most studies focused on above-ground traits, whereas the assessment of root trait diversity lags far behind (Barkaoui et al. 2016). One reason for that lies in the methodological difficulties to identify and separate roots in plant communities, in order to measure root traits at the individual level (Freschet et al. 2021a). To start filling this gap of knowledge, we used fine root diameter distribution at the community level as an indicator of root morphological diversity, which is one aspect of root functional diversity.

Fine roots (< 2 mm) diameter play a key role in plant (Ryser and Lambers 1995, Comas and Eissenstat 2004, Gregory 2006) and ecosystem functioning (Wardle et al. 1998, Birouste et al. 2012) due to their multifunctionality (Weemstra et al. 2016, Laliberté 2017, Freschet et al. 2021a). They have a hierarchical organization that consists in different root orders with varying diameters and functions. The most distal and thinner two or three root orders are involved in resource acquisition and uptake (so called 'absorptive roots', McCormack et al. 2015). The higher-order roots are coarser and mainly involved in water and nutrient transport (so called 'transport roots', McCormack et al. 2015). The hierarchical organization of the root system at the individual level typically translates into a skewed distribution of root length in different diameter classes, with either a single mode (unimodal distribution; Scanlan and Hinz 2010, Ryser et al. 2011, Caplan et al. 2019), two modes (bimodal distribution; Boot and Mensink 1990, Eissenstat 1991, Ryser and Lambers 1995, Bouma et al. 2000, Anderson et al. 2007, Roumet et al. 2016, Bodner et al. 2019), three modes or more (multimodal distribution; Blouin et al. 2007, Zobel et al. 2013, Roumet et al. 2016). The modes represent different root orders (Liu et al. 2018) or root types (absorptive versus transport; Fig. 1A). The first to third root orders that represent the absorptive roots usually group in one mode with a low average root diameter (M1), whereas the higher root orders, which represent the transport roots, group in one or several modes that show larger average root diameters (M2; Fig. 1A). Several studies have attempted to define metrics characterizing root diameter distribution at the individual level, and considered either the whole distribution (Caplan et al. 2019) or the various modes of the distribution (Anderson et al. 2007) in terms of shape, notably mean (M), standard deviation (SD) and relative proportions (P) of individual modes (Anderson et al. 2007; Fig. 1).

The root diameter distribution differs across life forms (Scanlan and Hinz 2010, Freschet et al. 2017, Ma et al. 2018) and species (Roumet et al. 2016, Caplan et al. 2019, Bodner et al. 2019). For example, grass species with a herringbone branching system and no secondary growth usually have a unimodal distribution (Fig. 1A) with a low mean value (M1, Fig. 1B), because most root length is composed of very thin and short-lived absorptive roots (McCormack et al. 2015), which allow dynamic exploration of the soil volume. Conversely, forbs, shrubs and trees show a more dichotomous branching system with several root orders and usually show a bimodal distribution representative of absorptive and transport roots, respectively (Fig. 1A, Liu et al. 2018). For woody species, the lower order and thinner roots usually forms a dominant mode (high P1) reflecting the absorptive roots, and the higher order and coarser root modes show a lower relative proportion (P2), and reflect transport roots (Wahl and Ryser 2000; Fig. 1A–B). In addition, the mean value (M1′) of the absorptive root mode is generally higher than in grass species (M1′ > M1), which reflects coarser and longer-lived absorptive roots (Fitter et al. 2002, Shao et al. 2019; Fig. 1B) foraging resources in a given soil volume over a longer period.

At the species level, the distribution of root diameter can also reflect plastic adaptations along a gradient of abiotic environmental filtering (Fig. 1C). For example, a shift toward higher diameters was observed in response to increased nutrient availability for grass species (M1'' > M1, Ryser and Lambers 1995). In the same vein, several studies have tested changes in root system morphology of a given species in response to 1) abiotic filters, such as water and nutrient availability (Kuchenbuch et al. 2006) and soil texture (Anderson et al. 2007), 2) disturbances, such as tillage (Pagliai and Nobili 1993, Qin et al. 2004) and 3) biotic changes, such as defoliation (Anderson et al. 2007). However, no study has addressed changes in root diameter distribution at the community level in response to changing environmental filtering. More generally, no study has addressed whether the diameter modes reflecting a functional hierarchy within species would translate to analogous differentiation and multimodality at the community level.

The root diameter distribution within a soil sample taken from a plant community should provide an integrative picture of the coexistence of root types and resource exploration strategies, encompassing intra-individual, intraspecific and interspecific variations within the community. First, we posit that the existence of separable modes should reveal a functional differentiation of roots at community-level. In particular, we expected that unimodal distributions reflect lower root morphological diversity and the predominance of one root morphological type. By contrast, multi-modal distributions should indicate a higher root morphological diversity that reflect a combination of several root types and a greater niche partitioning (Fig. 1D). Second, we expect that the width of a mode reflects the diversity of root strategies within each root mode. The narrower it is, the most constrained is the variability within the given root type. Third, we posit that the relative importance of the modes (P) inform about the relative importance of the different root types at the community level (Fig. 1E).

Our objective was to investigate root diameter distributions at the community level in varying environmental conditions to address how it captures the root morphological diversity stemming from plant assembly dynamics. We tested

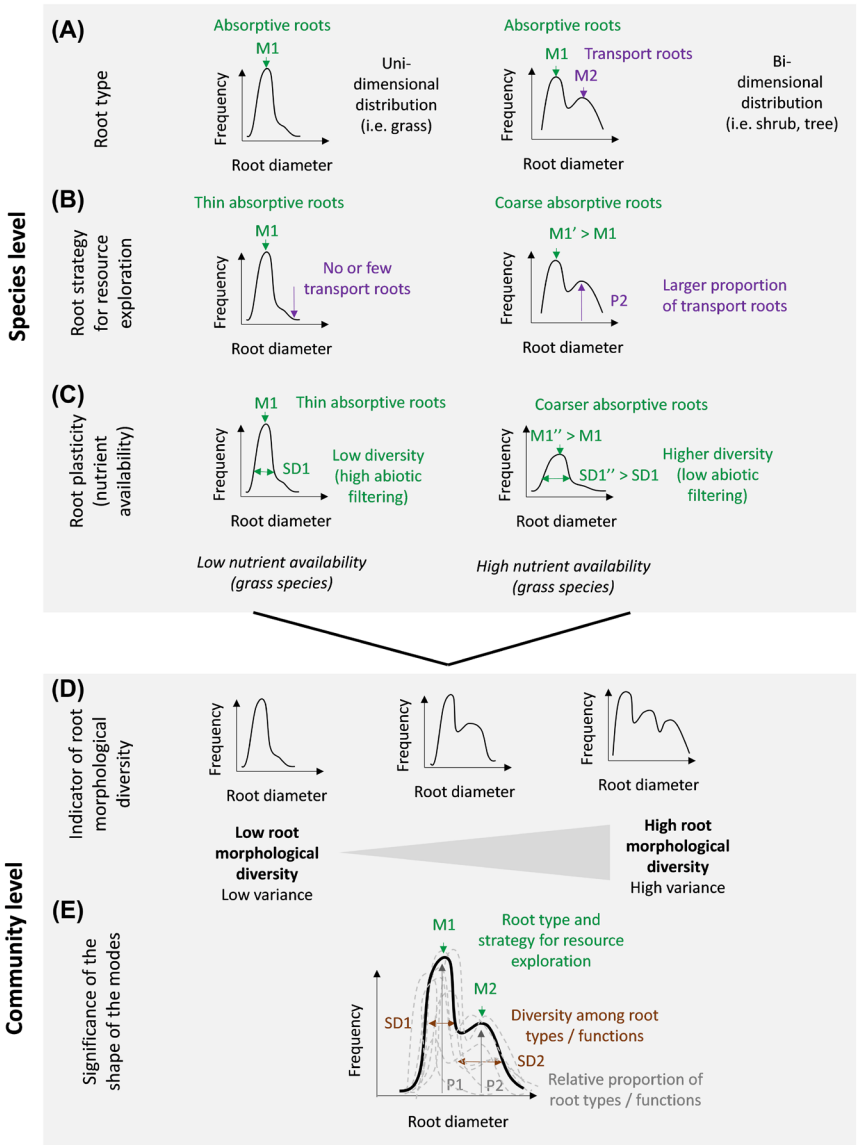

Figure 1. Overview of the insights brought by the study of root diameter distribution at the species level and how it can be upscaled to the community level. Mx, SDx and Px are the average root diameter, the standard deviation and the relative proportion of the mode x, respectively. M1′ and M1″ indicate variations in the average value of M1 related to interspecific variations (M1′) or plasticity (M1″). In the lower panel, dotted lines represents the fine root diameter distribution of the individual species that make up the model community, and the bold continuous black line represents the fine root diameter distribution of the resulting model community.

changes in community-level root diameter distribution along a successional gradient with and without yearly mowing, and assessed how the variations in parameters of root distribution could relate to vegetation composition and other ecological properties at community level. Whereas directional changes in average root traits are known (Holdaway et al. 2011, Erktan et al. 2018, Caplan et al. 2019), very little is known about root diversity and complementarity in root types and exploration strategies as succession proceeds. Usually, successional gradients are characterized by a decrease in nutrient availability and an increase in competition for resources (Tilman 1985), associated with the replacement of ruderal species by stress-tolerant and/or competitive species (Navas et al. 2010). Along the succession, average root traits points a shift from fine roots characterized by low average root diameter, high specific root length, low root dry matter content and low root carbon content that suggest a dynamic exploration of soils resources, to roots with opposite trait syndromes (Erktan et al. 2018, Caplan et al. 2019). We expected that root morphological diversity should increase along the succession due to the replacement of ruderal species by a mixture of stress-tolerant and competitive species with contrasting root morphologies. Finally, previous studies showed a limited effect of annual mowing on average root traits at the community level (Erktan et al. 2018), but in some cases (Caplan et al. 2019), mowing allowed to maintain ruderal

species in later successional stages. As such, we expected that mowing increases root morphological diversity in late successional stages leading to a multimodal root diameter distribution.

Material and methods

Study site

We sampled vegetation on 24 roadsides (plot dimension = 16 × 4 m) along a chronosequence (6–69 year-old) in the Mediterranean region, north of Montpellier (43°6′N, 3°8′E), southern France (mean annual temperature: 14.6 ± 6.80°C; mean annual precipitation: 774 ± 210 mm, INRA, Agroclim, France). On each roadside, a plant community (1.5–3 m wide strip) was yearly mown (8–10 cm height) and the adjacent community was left unmown, hence making n = 48 plant communities in total. Plots were all located on calcareous substrate with shallow (10–15 cm deep soil over bedrock in average) and silt dominated (39 ± 9%) soil. Changes in soil properties along the succession mainly concerned soil organic matter accumulation, with significant increase in soil organic carbon (SOC; Erktan et al. 2018). The percentage cover of plant growth forms in communities changed along the successional gradient (Erktan et al. 2018), with decreasing herbaceous cover and increasing shrub and tree covers ($p < 0.01$, Erktan et al. 2018), while graminoid cover did not vary significantly along the succession (Erktan et al. 2018). Further details on plant community composition are provided in Bouchet et al. (2017), and the percentage cover and mycorrhizal type (taken from Soudzilovskaia et al. 2020) of the three main dominant plant species per plot are indicated in the Supporting information.

Measuring fine root diameter distribution and root traits

In each of the 48 plant communities, we sampled 10 soil cores (3 cm diameter × 10 cm depth) in mid-April 2014, at the peak of vegetative growth, and pooled them to obtain composite soil samples including a mixture of roots from the species co-occurring in each plant community. Shallow soils from Mediterranean roadsides limit root growth beyond 10–15 cm depth, so that we assumed that most of the roots were located in the shallow soil layer (0–10 cm), from which we harvested soil samples. We removed tap roots and rhizomes, and selected and washed only fine roots (diameter < 2 mm). A subsample of fine roots (ca 700 mg fresh mass), representative of all the fine roots harvested in the community, was stained in a methyl violet solution (2 min – 0.5 g l^{-1}), rinsed, spread out on a transparent sheet and scanned on a flatbed scanner at 1200 dpi (Hummel et al. 2007). We analyzed the root images using an image analysis software (WinRhizo Pro ver. 2009c) with automatic thresholding option (Bouma et al. 2010), to determine the root length (L) within 40 root diameter classes, from 0 to 2 mm, with a 0.05 mm increment. After scanning, roots from the subsample were immediately weighed to determine their fresh mass (FM), oven-dried at 60°C for 72 h and weighed to determine their dry mass (DM). The community-level specific root length (SRL, m g^{-1}) calculated as the ratio between L and DM, and community-level root dry matter content (RDMC; mg g^{-1}) calculated as the ratio between DM and FM were analyzed in Erktan et al. (2018). In addition, the root concentration in nitrogen (Root N) and carbon (Root C) were measured on dried (60°C for 72 h) and finely milled roots (< 2 mm) using an elemental analyser, and were also analyzed in Erktan et al. (2018).

Plant community functional richness and CSR proportions

We gathered functional trait information from the public TRY database (Kattge et al. 2020, request no. 14939), for leaf area (LA), leaf dry matter content (LDMC), specific leaf area (SLA), vegetative height (VH) and seed mass (SM). We calculated CSR (competitive, stress-tolerant, ruderal) ecological strategies of plant species by applying the method of Pierce et al. (2017). We calculated the functional space defined by SLA, VH and SM (Westoby 1998), each scaled to 0 mean and 1 variance. We calculated the functional richness (Fric) of each plant community in this functional space by means of the *dbFD* function of R package FD (Laliberté and Shipley 2012). Variations of plant community functional richness and in the relative proportions of competitive (C), stress-tolerant (S) and ruderal (R) strategies with age and mowing are given in the Supporting information.

Characterizing and analyzing root diameter distributions

We characterized the modes of root diameter distribution by fitting a Gaussian mixture model (*mixfit* function in R package mixR). The distribution was defined based on the proportion of root length within each root diameter class. Because the distribution is bounded by 0 and globally skewed, we fitted a lognormal mixture model. We determined whether one, two or three modes could be identified, by comparing and testing the likelihoods of models with 1–3 modes. The selected model yielded NA values for the second (respectively third) mode when no more than one (respectively two) mode(s) could be fitted. For each mode, we calculated the mean M, standard deviation SD and relative proportions P, as in Anderson et al. (2007). The parameter P quantified the relative proportion of the modes in the mixture model. The script to analyze the root diameter distribution is provided in the Supporting information.

Variation of root diameter distribution with succession and mowing

We inspected variations in root diameter distribution parameters along the succession and in response to mowing by using mixed models, with age, mowing and their interactions

as fixed effects, and roadside as random effect. Relationships between root diameter distributions parameters were tested using pairwise Spearman's correlation coefficient (ρ), and tested with Bonferroni correction. Similarly, we tested the correlation between the root diameter distribution parameters (M, SD, P, Variance), the community-level average fine root traits (mean root diameter, SRL, RDMC, Root N, Root C), and the characteristics of the plant community (plant community specific and functional richness, proportions of growth forms and CSR strategies) and of the soil chemistry (CEC, pH, soil available P, soil organic C and total soil N). The species diversity [Simpson], percentage of grass, herbs, shrubs and trees, woodiness (cover of woody species) and the soil characteristics were taken from Erktan et al. (2018). All statistical analyses were conducted with R ver. 4.0.3 (<www.r-project.org>).

Results

Variation and co-variation of the parameters of root diameter distributions

The root diameter distributions of plant communities were mainly multimodal. Out of the 48 communities, four showed a unimodal (Fig. 2A–B), 13 a bimodal (Fig. 2C–D) and 31 a

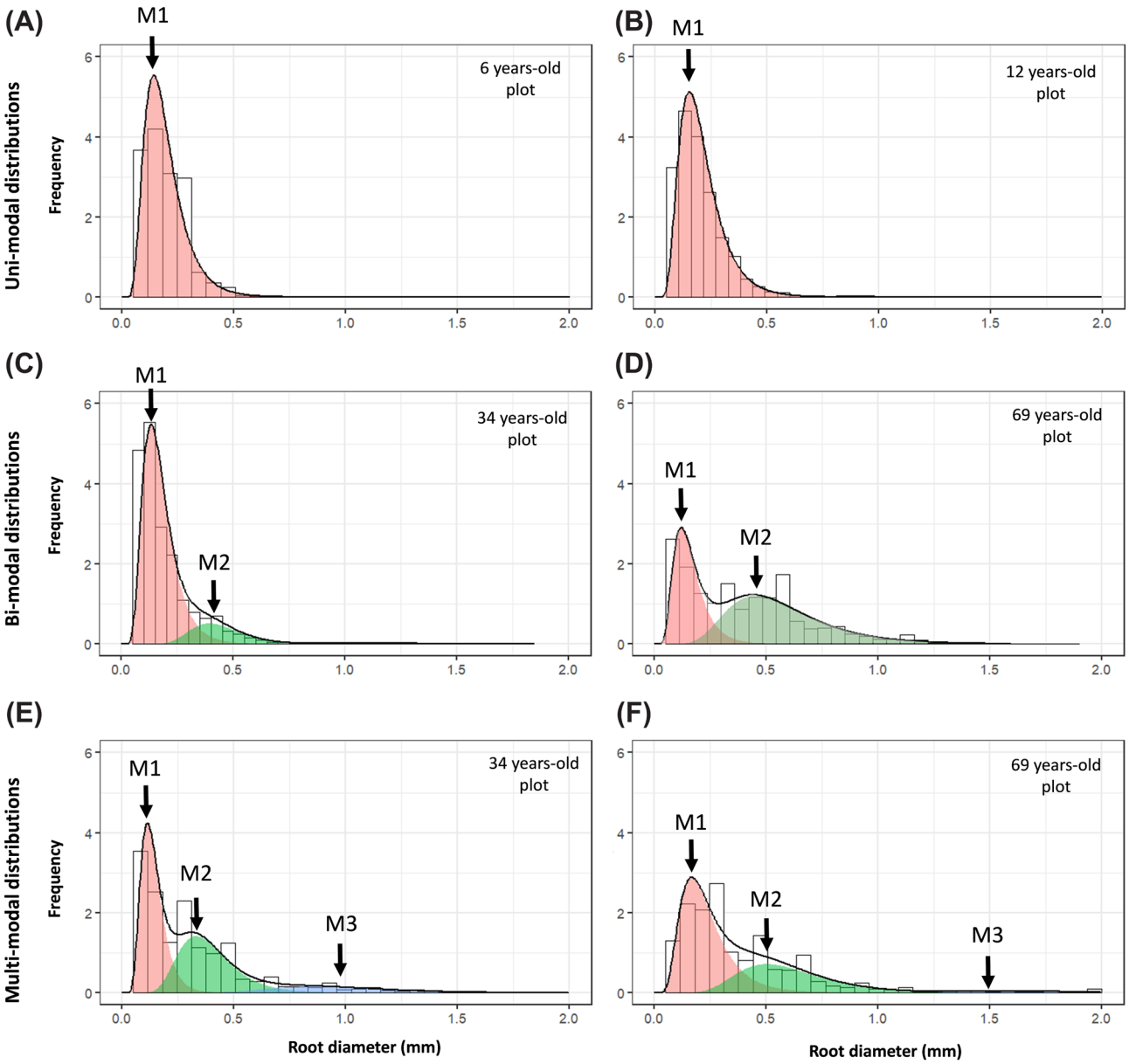

Figure 2. Examples of root diameter distributions of plant communities along the successional gradient. Uni- (A, B), bi- (C, D) and multi-modal (E, F) distributions are displayed. Black arrows indicates the different modes. Black lines indicate the fitted distribution of the model. Red, green and blue lines indicate the fitted distribution of the first, second and third modes, respectively.

trimodal (Fig. 2E–F) distribution. On average, the first mode represented the highest proportion of root length ($\overline{P1} = 0.8 \pm 0.2$), with thinner average diameter ($\overline{M1} = 0.21 \pm 0.06$ mm) and narrower width ($\overline{SD1} = 0.1 \pm 0.06$ mm). The second mode corresponded to larger roots ($\overline{M2} = 0.54 \pm 0.21$ mm), included most of the remaining proportion of root length ($\overline{P2} = 0.2 \pm 0.2$) and was wider ($\overline{SD2} = 0.17 \pm 0.07$ mm). The third mode included a much lower proportion of the total root length ($\overline{P3} = 0.02 \pm 0.002$), corresponded to markedly coarser roots ($\overline{M3} = 1.3 \pm 0.45$ mm) and was as wide ($\overline{SD3} = 0.15 \pm 0.09$ mm) as the second mode.

The parameters describing the root diameter distribution at the community level were poorly correlated, except for specific couples of variables. The mean value of the second mode (M2) was positively related to the mean value of the first (M1, $\rho = 0.56$; $p = 0.004$) and third mode (M3, $\rho = 0.59$; $p = 0.02$; Table 1). The widths of the three modes (SD1, SD2 and SD3) were not correlated to each other. However, the width of the first mode (SD1) increased together with the mean value of the first (M1, $\rho = 0.87$; $p < 0.001$) and of the second mode (M2, $\rho = 0.53$; $p = 0.01$). Similarly, the width of the second mode (SD2) was positively related to the mean value of the third mode (M3, $\rho = 0.67$; $p = 0.002$). The proportions of the two main modes (P1 and P2) were strongly negatively related ($\rho = -0.99$; $p < 0.001$). Finally, the overall variance of the root diameter distribution was positively related to the mean value of the third mode (M3, $\rho = 0.57$; $p = 0.04$) and to the proportion of the second mode (P2, $\rho = 0.48$; $p = 0.04$).

Influence of ecological succession and mowing on root diameter distributions

Along the succession, the mean root diameter of the first root mode (M1) increased significantly ($p = 0.01$), and root diameters within this mode showed more variability (increasing SD1; $p = 0.007$) (Fig. 3, Supporting information). Conversely, the mean root diameter in the two coarser modes (M2 and M3) remained unchanged, while the width of the third mode (SD3) increased along the succession ($p = 0.01$). The proportion of the first mode (P1) decreased ($p = 0.03$), whereas the proportion of second mode (P2) increased ($p = 0.002$), and the proportion of third mode (P3) remained low and unchanged (Fig. 3, Supporting information). The overall variance of the root diameter distribution significantly increased along the succession ($p = 0.001$). Mowing did not significantly affect any of the parameters of the root diameter distribution (Fig. 3, Supporting information).

We analyzed pairwise correlations between the distribution parameters, the community-level average root traits and the plant community and soil characteristics, in order to assess how the variations in parameters of root diameter distribution could relate to vegetation composition and other ecological properties at community level (Table 2). Overall, the first mode was not related to community characteristics (proportion of plant growth forms, CSR strategies and plant functional richness) but was related to morphological root traits. Specifically, the mean of the first mode (M1) increased with the mean root diameter of the plant community ($\rho = 0.45$, $p = 0.001$) and decreased with the specific root length (SRL; $\rho = -0.41$, $p = 0.04$). The width of the first mode (SD1) was positively correlated with the root dry matter content (RDMC; $\rho = 0.44$, $p = 0.02$). The second mode, in particular its proportion (P2), was mainly related to the percentage of woody species ($\rho = 0.53$; $p = 0.002$), including shrubs ($\rho = 0.50$; $p = 0.005$) and trees ($\rho = 0.51$; $p = 0.004$). We also found that P2 was positively related to the plant functional richness ($\rho = 0.44$; $p = 0.03$) and the proportion of stress tolerant species ($\rho = 0.48$, $p = 0.01$). Conversely, P2 was negatively related to the SRL ($\rho = -0.55$, $p = 0.001$) and the proportion of ruderal species ($\rho = -0.55$, $p = 0.001$). The width of the second mode (SD2) was positively related

Table 1. Spearman correlation (ρ) matrix among root distribution parameters (n = 48). Displayed are Bonferroni adjusted p-values. Bold characters indicate significant correlations.

	M2	M3	SD1	SD2	SD3	P1	P2	P3	Variance
M1	**0.56** **p = 0.004**	0.42 p = 0.92	**0.87** **p < 0.001**	0.05 p = 1.00	−0.16 p = 1.00	0.35 p = 0.62	−0.29 p = 1.00	0.02 p = 1.00	0.32 p = 1.00
M2		**0.59** **p = 0.02**	**0.53** **p = 0.01**	0.38 p = 0.54	0.02 p = 1.00	0.42 p = 0.20	−0.41 p = 0.28	−0.22 p = 1.00	0.38 p = 0.52
M3			0.38 p = 1.00	**0.67** **p = 0.002**	−0.18 p = 1.00	−0.03 p = 1.00	0.07 p = 1.00	−0.21 p = 1.00	**0.57** **p = 0.04**
SD1				0.02 p = 1.00	−0.10 p = 1.00	0.40 p = 0.20	−0.26 p = 1.00	0.02 p = 1.00	0.32 p = 1.00
SD2					−0.26 p = 1.00	−0.29 p = 1.00	0.34 p = 1.00	−0.06 p = 1.00	0.47 p = 0.07
SD3						0.06 p = 1.00	−0.11 p = 1.00	0.52 p = 0.10	0.21 p = 1.00
P1							**−0.99** **p < 0.001**	−0.56 p = 0.06	−0.33 p = 0.88
P2								0.48 p = 0.29	**0.48** **p = 0.04**
P3									0.47 p = 0.34

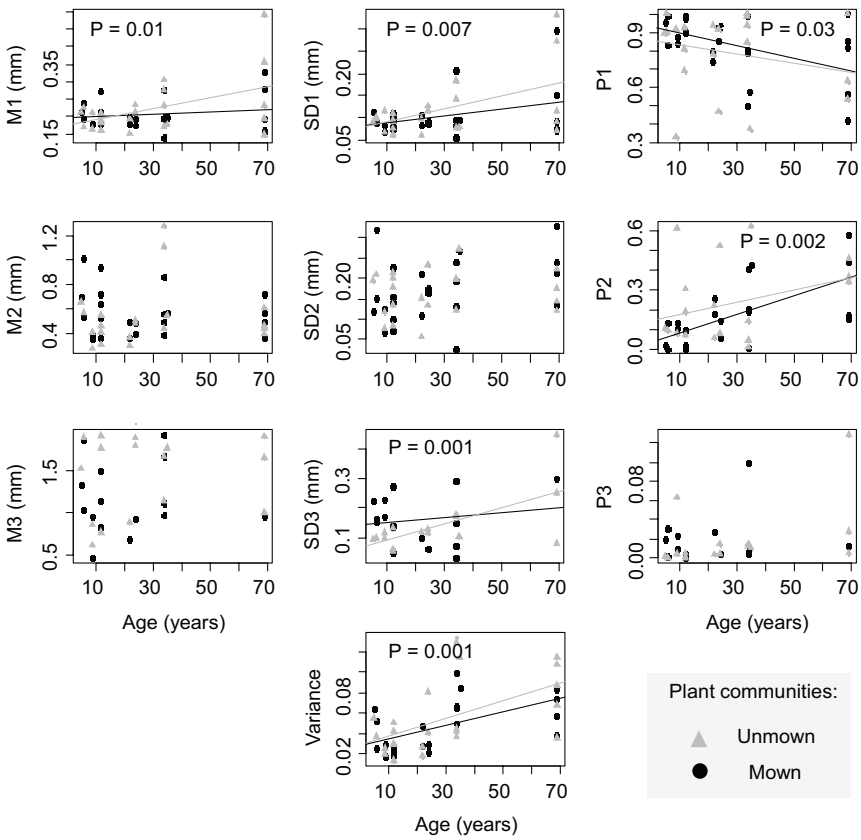

Figure 3. Variations in root diameter distribution parameters along the succession gradient and in response to mowing. Black circles represent mown communities and grey triangles represent unmown communities. The lines represent predictions of mixed linear models (black = mown communities; grey = unmown communities). Mx; SDx and Px are the average root diameter, the standard deviation and the relative proportion of the mode x, respectively. Variance represents the overall variance of the entire root diameter distribution. The p-values indicate the significance of the age effect in mown and unmown communities together. Only significant effects are displayed. More information about the models is provided in the Supporting information.

to the plant community functional richness ($\rho = 0.43$, $p = 0.03$), and negatively related to the SRL of the entire plant community ($\rho = -0.42$, $p = 0.04$). The mean value of the third mode (M3) was positively related to root C concentration ($\rho = 0.59$, $p = 0.005$) and negatively associated with the SRL ($\rho = -0.58$, $p = 0.007$).

The variance of the root diameter distribution was strongly related to several community-level average root trait values and plant community characteristics. In particular, the variance of the root diameter distribution was positively correlated to the mean root diameter ($\rho = 0.72$, $p < 0.001$), the RDMC ($\rho = 0.60$, $p < 0.001$), the root C ($\rho = 0.70$, $p < 0.001$), and the proportion of shrubs ($\rho = 0.41$, $p = 0.04$), trees ($\rho = 0.63$, $p < 0.001$) and woody species ($\rho = 0.57$, $p < 0.001$). Furthermore, it was positively related to the functional richness of the plant communities ($\rho = 0.54$, $p = 0.001$), and to the proportions of competitive ($\rho = 0.55$, $p < 0.001$) and stress-tolerant species ($\rho = 0.44$, $p = 0.02$). On the contrary, the variance of the root diameter distribution was negatively related to the SRL ($\rho = -0.93$, $p < 0.001$) and to a lesser extent to the proportion of ruderal species ($\rho = -0.44$, $p = 0.02$) and species diversity (Simpson, $\rho = -0.42$, $p = 0.03$).

The parameters describing the root diameter distribution and the soil characteristics were poorly related. Neither the CEC, nor the soil P related to any of the root distribution parameters. Only the concentration in soil organic C and in total N were positively related to the proportion of the second mode (P2, $\rho = 0.48$, $p = 0.01$ and $\rho = 0.49$, $p = 0.007$, respectively) and to the overall variance of the distribution ($\rho = 0.57$, $p < 0.001$ and $\rho = 0.55$, $p < 0.001$, respectively). In addition, the soil pH was negatively associated with the variance of the distribution ($\rho = -0.55$, $p < 0.001$).

Discussion

Ecological meaning of the modes in root diameter distribution at the plant community level

Most communities showed a multimodal distribution of root diameters, with two main modes, and a third mode with much lower importance. The multimodularity of the root distribution could result from the intraspecific variation of root diameter within a fine root branching hierarchy (Lecompte et al. 2005), and from the interspecific variation of

Table 2. Spearman correlation (ρ) matrix between root distribution parameters, average root traits and plant community and soil characteristics (n = 48). Displayed are Bonferroni adjusted p-values. Bold characters indicate significant correlations. MRD: mean root diameter (plant community level); Plant Fric: plant functional richness; % C: percentage of competitive species; % S: percentage of stress-tolerant species; % R: percentage of ruderal species; CEC: cation exchange capacity.

	Fine root diameter distribution parameters									
	M1	SD1	P1	M2	SD2	P2	M3	SD3	P3	Variance
Community level average root traits										
MRD	**0.45**	0.36	−0.25	0.35	0.32	0.37	0.39	0.02	0.43	**0.72**
	p = 0.001	p = 0.1	p = 0.9	p = 0.20	p = 0.3	p = 0.1	p = 0.3	p = 1.00	p = 0.2	**p < 0.001**
SRL	**−0.41**	−0.40	0.38	−0.33	**−0.42**	**−0.55**	**−0.58**	−0.09	−0.46	**−0.93**
	p = 0.04	p = 0.05	p = 0.08	p = 0.30	**p = 0.04**	**p = 0.001**	**p = 0.007**	p = 1.00	p = 0.10	**p < 0.001**
RDMC	0.32	**0.44**	−0.09	0.24	0.27	0.26	0.44	0.19	0.18	**0.60**
	p = 0.30	**p = 0.02**	P = 1.00	p = 1.00	p = 0.72	p = 0.84	p = 0.15	p = 1.00	p = 1.00	**p < 0.001**
Root N	−0.25	−0.29	0.14	−0.19	0.05	−0.12	−0.02	0.14	−0.10	−0.16
	p = 0.80	p = 0.41	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00
Root C	0.31	0.31	−0.28	0.22	0.37	0.38	**0.59**	0.19	0.28	**0.70**
	p = 0.40	p = 0.33	p = 0.50	p = 1.00	p = 0.13	p = 0.10	**p = 0.005**	p = 1.00	p = 1.00	**p < 0.001**
Plant community characteristics										
Simpson	−0.23	−0.31	0.02	−0.017	0.11	−0.20	−0.13	−0.34	−0.42	**−0.42**
	p = 1.00	p = 0.34	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 0.63	p = 0.20	**p = 0.03**
Grass (%)	−0.08	−0.14	0.001	0.22	0.04	−0.07	−0.06	−0.08	−0.14	−0.08
	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00
Herbs (%)	−0.15	−0.17	0.20	0.05	−0.06	−0.35	−0.15	0.019	−0.24	**−0.40**
	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 0.21	p = 1.00	p = 1.00	p = 1.00	**p = 0.04**
Shrubs (%)	0.17	0.15	−0.36	0.002	0.31	**0.50**	0.23	−0.09	0.19	**0.41**
	p = 1.00	p = 1.00	p = 0.12	p = 1.00	p = 0.42	**p = 0.005**	p = 1.00	p = 1.00	p = 1.00	**p = 0.04**
Trees (%)	0.11	0.14	−0.30	−0.02	0.44	**0.51**	0.43	0.14	0.31	**0.63**
	p = 1.00	p = 1.00	p = 0.39	p = 1.00	p = 0.02	**p = 0.004**	p = 0.15	p = 1.00	p = 0.93	**p < 0.001**
Woodiness (%)	0.20	0.21	−0.33	−0.10	0.28	**0.53**	0.33	−0.02	0.27	**0.57**
	p = 1.00	p = 1.00	p = 0.22	p = 1.00	p = 0.61	**p = 0.002**	p = 0.70	p = 1.00	p = 1.00	**p < 0.001**
Plant Fric	0.26	0.30	−0.31	0.13	**0.43**	**0.44**	0.42	−0.14	0.16	**0.54**
	p = 0.71	p = 0.38	p = 0.30	p = 1.00	**p = 0.03**	**p = 0.03**	p = 0.19	p = 1.00	p = 1.00	**p = 0.001**
% C	0.04	0.11	−0.08	0.27	0.16	0.23	0.18	0.24	0.22	**0.55**
	p = 1.00	p = 1.00	p = 1.00	p = 0.71	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	**p < 0.001**
% S	0.26	0.25	−0.27	0.07	0.37	**0.48**	0.23	0.12	0.37	**0.44**
	p = 0.71	p = 0.81	p = 0.62	p = 1.00	p = 0.14	**p = 0.01**	p = 1.00	p = 1.00	p = 0.38	**p = 0.02**
% R	−0.25	−0.29	0.34	0.09	−0.35	**−0.55**	−0.33	0.22	−0.08	**−0.44**
	p = 0.81	p = 0.45	p = 0.17	p = 1.00	p = 0.21	**p = 0.001**	p = 0.74	p = 1.00	p = 1.00	**p = 0.02**
Soil characteristics										
Soil CEC	−0.16	−0.04	−0.12	0.06	0.09	0.18	0.10	0.01	0.02	0.19
	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00
Soil pH	−0.06	−0.05	0.27	−0.03	−0.19	−0.4	−0.2	0.06	−0.23	**−0.55**
	p = 1.00	p = 1.00	p = 0.58	p = 1.00	p = 1.00	p = 0.02	p = 1.00	p = 1.00	p = 1.00	**p < 0.001**
Soil P (Olsen)	−0.16	−0.12	0.08	−0.10	−0.18	−0.11	−0.03	−0.17	−0.01	−0.11
	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00	p = 1.00
Soil C org	0.09	0.14	−0.30	0.04	0.31	**0.48**	0.24	−0.07	0.23	**0.57**
	p = 1.00	p = 1.00	p = 0.37	p = 1.00	p = 0.42	**p = 0.01**	p = 1.00	p = 1.00	p = 1.00	**p < 0.001**
Soil N (total)		0.03	−0.34	−0.06	0.20	**0.49**	0.21	0.06	0.32	**0.55**
		p = 1.00	p = 0.20	p = 1.00	p = 1.00	**p = 0.007**	p = 1.00	p = 1.00	p = 0.78	**p < 0.001**

root diameter for both absorptive and transport roots (Fig. 1, McCormack et al. 2015). The fact that we observe modes at the community level indicate that the combination of root morphologies from different species maintain that some root diameter values are found in higher frequencies than others, otherwise a random distribution would have been observed. From a methodological point of view, the multimodal distribution of root diameters questions the wide use of community-level average root traits (Holdaway et al. 2011, Roumet et al. 2016, Bergmann et al. 2020), notably the mean root diameter.

The shape of the modes provides complementary insights on the different root types within the pool of fine roots < 2 mm. The first mode could reflect the properties of the most absorptive roots at community level, as this mode represented most of the root length ($\overline{P1} = 0.8 \pm 0.2$) and the thinnest roots ($\overline{M1} = 0.21 \pm 0.06$ mm), which are usually at more distal root ends and ensure the absorption of water and nutrients (McCormack et al. 2015). Investigating the relationship between distribution parameters and community-level roots trait values showed that M1 was unrelated to the proportions of plant growth forms and the CSR strategies, suggesting

that the first mode represents absorptive roots of a mixture of coexisting plant species.

The second mode included larger-diameter roots ($\overline{M2} = 0.54 \pm 0.21$ mm), which could belong to woody species as suggested by the positive relation between the proportion of woody species, namely shrubs and trees and P2 (Table 2). A large part of the roots of the second mode could be transport roots, as they are generally thicker and typical of the dichotomous woody root systems (McCormack et al. 2015). Specifically, a study on 65 tree species pointed that the mean diameter of the first-order root ranged from 0.25 to 0.38 mm (Chen et al. 2013), thereby below $\overline{M2}$. Similarly, in two recent meta-analyses, the mean diameter of first order root from woody species were 0.28 mm (Wang et al. 2018) and 0.30 mm (visual estimation from the graph provided in Ma et al. 2018), respectively. In the same vein, a rapid investigation of the average diameter of fine roots from the dominant shrub and tree species of our case study (14 species found) in the root trait database GRooT (Guerrero-Ramirez et al. 2020) showed that they were on average finer (0.44 mm) than $\overline{M2}$. Only the highest values (0.55 mm) corresponding to *Quercus ilex* were close to $\overline{M2}$. Altogether, these arguments suggest that the second root mode is mainly composed by transport roots from woody species, whereas woody absorptive roots are mostly included in the first mode.

The third mode represented a far smaller proportion of root length ($\overline{P3} = 0.02 \pm 0.002$) and concerned much coarser roots ($\overline{M3} = 1.3 \pm 0.45$ mm), although present in many communities (31 over 48). The positive relationship between $\overline{M3}$ and the total root C suggests that the roots contributing to this mode present a higher proportion of structural tissues. As a consequence, the third root mode presumably represents large transport roots. The 2 mm diameter cut-off used for the initial root sorting (as often used in root ecology, McCormack et al. 2015) did not allow to consider the entire root diameter distribution. As a consequence, coarser transport roots could be omitted. Further investigation of a wider root diameter distribution beyond the 2 mm cut-off should allow better capturing the contribution of these roots. To sum up, our results support that morphological differentiation of modes in root diameter distribution can reflect functional differences among absorptive and transport roots at the community level.

Variations along the succession and in response to mowing

The overall variance of the root diameter distribution increased along the succession, reflecting an increase in root morphological diversity. The positive relationship between this variance and the plant functional richness (based on above-ground traits), but not with plant species diversity, suggests that the variance of the root diameter distribution is related to global functional variation within communities. Moreover, the variance of the root distribution was positively related to the proportion of competitive and stress-tolerant species and negatively to the proportion of ruderal species, suggesting that the coexistence of the latter strategies in the older sites involved greater functional complementarity between root types and strategies, while dynamic explorative strategies displayed by ruderal plants could predominate in early succession.

The variation of mode parameter values along the succession reflected the influence of successional dynamics on root morphological diversity. The diameter of the first root mode (M1) increased in later succession, which suggested a shift in soil resource exploration strategy. Thinner roots have indeed an inherently greater capacity to encountered soil resources based on higher surface area per unit of mass, and have a high root turnover (McCormack et al. 2012); they thus have a high dynamic explorative strategy in the early successional sites. In later successional stages, the thicker roots are traditionally assumed to have a more conservative strategy because they are long-lived (Tjoelker et al. 2005, Reich 2014, Weemstra et al. 2016). Recent studies demonstrated that thick roots are also associated with a strong potential to host mycorrhizal partners and thus could explore soil very efficiently via their hyphae network (outsourcing strategy) (McCormack and Iversen 2019, Bergmann et al. 2020). This is consistent with previous studies showing that mycorrhizal colonization increased with successional age (Hartnett and Wilson 1999, Rasmann et al. 2011, Tedersoo et al. 2020). In addition, the first mode showed a broader range of root diameters (SD1) as succession proceeded, suggesting a coexistence of more diverse root absorptive strategies in later succession, including thin roots with highly dynamic explorative strategy, mainly from remaining grass and herbs, and an increasing proportion of absorptive thicker roots with more conservative and/or outsourcing strategy, from woody species. Finally, the relative proportion of root length in the first mode (P1) significantly decreased with age, but still remained important in late succession (Fig. 3), suggesting that absorptive roots remained abundant in communities (in terms of proportion in root length) throughout the succession. Remarkably, the decrease in P1 was not related to the reduced abundance of grass and herb species along the succession (Table 2), which reflect that the absorptive roots in the first mode did not only belong to ruderal species.

The relative proportion of the second mode (P2) changed and increased along the succession, at the expense of P1. Higher P2 value suggested a higher proportion of transport roots in later succession, which was associated to a higher proportion of woody species (Erktan et al. 2018) and stress-tolerant species (Table 2, Supporting information). In addition, although SD2 did not vary along the succession, its positive relationship with plant community functional richness could reflect the coexistence of transport roots with contrasting dimensions, plausibly belonging to different plant CSR strategies.

Only the width of the third mode (SD3) increased along the succession. This increase was related neither to any plant community and soil characteristics, nor to community level average root traits. The increase in SD3 with age could reflect

greater contribution of older and longer-lived plant individuals in late succession, with larger-diameter transport roots, but which could still co-exist with younger and shorter-lived individuals with lower-diameter transport roots.

Although the parameters of the root diameter distribution strongly varied along the succession, they did not in response to mowing. This result did not confirm our hypothesis but is consistent with a globally low influence of mowing on community-level average root traits, notably due to the low mowing pressure that allow short-statured woody species to persist (Erktan et al. 2018).

Overall, inspecting the root diameter distributions of fine roots < 2 mm provided insights into the shifts and co-existence of root types and strategies for soil resources exploration, while community-level root trait analyses usually focus on the shifts in average root traits (Holdaway et al. 2011, Erktan et al. 2018, Caplan et al. 2019).

Root diameter distribution as an integrative measure of root morphological diversity at community level

Inspecting the root diameter distribution provides a low-cost and time-efficient way to assess root morphological diversity at the community level, which is one facet of root functional diversity. The parameters describing the shape of the distribution were little related to each other, supporting that they could reflect different and complementary facets of root morphological diversity. Nonetheless, a wider SD2 was related to a coarser third mode (greater M3), and the shifts of M1 and M2 were associated with shifts of the following modes M2 and M3, respectively. These relations could reflect concomitant changes in the diameter of absorptive and transport roots along the succession. While the variance of the entire root distribution covaried with the mean root diameter and SRL (average root traits) along the succession, the mode-specific parameters were broadly independent from average root traits and thus provided non-redundant information.

To analyze more finely variations in root diameter distribution in relation to root strategies for resource use and exploration, one may separate manually absorptive and transport roots before scanning as it is now generally recommended for root ecology studies (Freschet et al. 2021b), and analyze separately their root diameter distribution. In addition, root morphological diversity is only one facet of root functional diversity and there is an urgent need in further studies to be better accounted for root physiological traits (for example nutrient and water uptake capacity, mycorrhizal colonization, exudation) that determine root ability to acquire nutrients. In addition, phylogeny is increasingly recognized as an important determinant of root morphology (Valverde-Barrantes et al. 2017) and should be further considered to assess root functional diversity.

Another way to assess root functional diversity at the community level is to use root trait databases combined with plant community composition. To do so, one needs to identify plant species aboveground, then measure or take their average root traits values in databases (Iversen et al. 2017, Guerrero-Ramirez et al. 2020), and finally weight them using their relative abundance aboveground (Holdaway et al. 2011, Kramer-Walter et al. 2016, Caplan et al. 2019). With this approach, average root traits and functional diversity indices (variance, skewness and kurtosis) can be calculated. However, there are some disadvantages. First, root trait databases are still in their infancy, and data of many species and 'hard traits', notably related to root physiology, are still not available. For example, in the GRooT database (Guerrero-Ramirez et al. 2020) the root diameter was available only for 26 species among the 57 most abundant species found along the successional gradient. Second, this approach fails to account for local intraspecific variability. Third, it assumes that the abundance of species aboveground is a proxy of the belowground contribution of species in terms of root length, which is a debatable assumption since the root mass fraction strongly differ among species (Poorter et al. 2012, 2015, Ottaviani et al. 2020) and with environmental conditions (Poorter et al. 2012). Conversely, the root diameter distribution in soil cores bypasses the extrapolation of aboveground abundance to the belowground parts of the plants. Moreover, the root diameter distribution in soil cores synthesizes the functional variation within the community beyond the inter- and intraspecific sources of variation. It encompasses all species found in the soil core and does not require assigning each piece of roots to specific species.

Conclusion

We demonstrated that the fine-root diameter distribution synthesizes fine-root morphological diversity at a community level, and that the shape of its modes reveals distinctive root types and strategies for resource use and exploration. Most communities showed a multimodal distribution, with two main modes corresponding to absorptive roots (thinner root mode) and transport roots (coarser root mode). As succession proceeds, the relative proportion of absorptive roots (P1) decreased while those of transport roots (P2) increased mainly due to an increased proportion of woody species. Within absorptive roots, the average diameter value (M1) shifted from thinner absorptive roots allowing more explorative strategy in earlier succession, to thicker absorptive roots allowing a more conservative and/or outsourcing strategy in late successional stages. Considering the fine-root diameter distribution at the community level provides a low-cost and easy-implementable way to capture morphological diversity and complements the analysis of community average root trait values. As a consequence, it appears as an efficient starting point to study root morphological diversity at the community level, and would need to be further associated to root physiology and phylogeny to fully capture root functional diversity.

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Author contributions

Amandine Erktan: Conceptualization (equal); Formal analysis (lead); Methodology (equal); Writing – original draft (lead); Writing – review and editing (lead). **Catherine Roumet**: Conceptualization (equal); Formal analysis (supporting); Methodology (equal); Writing – original draft (supporting); Writing – review and editing (supporting). **François Munoz**: Conceptualization (equal); Formal analysis (supporting); Methodology (equal); Writing – original draft (supporting); Writing – review and editing (supporting).

Data availability statement

Data are available from the Dryad Digital Repository: <https://doi.org/10.5061/dryad.qv9s4mwgb> (Erktan et al. 2022).

Supporting information

The supporting information associated with this article is available from the online version.

References

Anderson, T. M. et al. 2007. Bimodal root diameter distributions in Serengeti grasses exhibit plasticity in response to defoliation and soil texture: implications for nitrogen uptake. – Funct. Ecol. 21: 50–60.
Barkaoui, K. et al. 2016. Mean root trait more than root trait diversity determines drought resilience in native and cultivated Mediterranean grass mixtures. – Agricult. Ecosyst. Environ. 231: 122–132.
Bergmann, J. et al. 2020. The fungal collaboration gradient dominates the root economics space in plants. – Sci. Adv. 6: eaba3756.
Birouste, M. et al. 2012. Plant traits and decomposition: are the relationships for roots comparable to those for leaves? – Ann. Bot. 109: 463–472.
Blouin, M. et al. 2007. A quick method to determine root biomass distribution in diameter classes. – Plant Soil 290: 371–381.
Bodner, G. et al. 2019. Characterization of cover crop rooting types from integration of rhizobox imaging and root atlas information. – Plants 8: 514.
Boot, R. and Mensink, M. 1990. Size and morphology of root systems of perennial grasses from contrasting habitatsnas affected nitrogen supply. – Plant Soil 129: 291–299.
Bouchet, D. et al. 2017. Mowing influences community-level variation in resource-use strategies and flowering phenology along an ecological succession on Mediterranean road slopes. – Appl. Veg. Sci. 20: 376–387.
Bouma, T. et al. 2000. Sample preparation and scanning protocol for computerised analysis of root length and diameter. – Plant Soil 218: 185–196.
Bouma, T. J. et al. 2010. Comparing ecosystem engineering efficiency of two plant species with contrasting growth strategies. – Ecology 91: 2696–2704.
Bruelheide, H. et al. 2018. Global trait–environment relationships of plant communities. – Nat. Ecol. Evol. 2: 1906–1917.
Buzzard, V. et al. 2016. Re-growing a tropical dry forest: functional plant trait composition and community assembly during succession. – Funct. Ecol. 30: 1006–1013.
Caplan, J. S. et al. 2019. Fine-root traits are linked to species dynamics in a successional plant community. – Ecology 100: e02588.
Chen, W. et al. 2013. Root trait variation and evolution. – Global Ecol. Biogeogr. 22: 846–856.
Chen, W. et al. 2016. Root morphology and mycorrhizal symbioses together shape nutrient foraging strategies of temperate trees. – Proc. Natl Acad. Sci. USA 113: 8741–8746.
Comas, L. H. and Eissenstat, D. M. 2004. Linking fine root traits to maximum potential growth rate among 11 mature temperate tree species. – Funct. Ecol. 18: 388–397.
Eissenstat, D. 1991. On the relationship between specific root length and the rate of root proliferation: a field study using citrus rootstocks. – New Phytol. 118: 63–68.
Erktan, A. et al. 2018. Two dimensions define the variation of fine root traits across plant communities under the joint influence of ecological succession and annual mowing. – J. Ecol. 106: 2031–2042.
Erktan, A. et al. 2022. Data from: Root traits, root diameter distribution and soil parameters at the community level along a mediterranean successional gradient. – Dryad Digital Repository, <https://doi.org/10.5061/dryad.qv9s4mwgb>.
Fitter, A. et al. 2002. Root system architecture determines fitness in an *Arabidopsis* mutant in competition for immobile phosphate ions but not for nitrate ions. – Proc. R. Soc. B 1504: 2017–2022.
Freschet, G. T. et al. 2017. Climate, soil and plant functional types as drivers of global fine-root trait variation. – J. Ecol. 105: 1182–1196.
Freschet, G. T. et al. 2021a. A starting guide to root ecology: strengthening ecological concepts and standardizing root classification, sampling, processing and trait measurements. – New Phytol. 232: 973–1122.
Freschet, G. T. et al. 2021b. Root traits as drivers of plant and ecosystem functioning: current understanding, pitfalls and future research needs. – New Phytol. 232: 1123–1158.
Garnier, E. et al. 2004. Plant functional markers capture ecosystem properties during secondary succession. – Ecology 85: 2630–2637.
Gregory, P. J. 2006. Plant roots: growth, activity and interactions with the soil. – Blackwell Publishing.
Gross, N. et al. 2007. Complementarity as a mechanism of coexistence between functional groups of grasses. – J. Ecol. 95: 1296–1305.
Guerrero-Ramirez, N. et al. 2020. Global root traits (GRooT) database. – Global Ecol. Biogeogr. 30: 25–37.
Hartnett, D. C. and Wilson, G. W. 1999. Mycorrhizae influence plant community structure and diversity in tallgrass prairie. – Ecology 80: 1187–1195.
Holdaway, R. J. et al. 2011. Species-and community-level patterns in fine root traits along a 120 000-year soil chronosequence in temperate rain forest. – J. Ecol. 99: 954–963.

Hummel, I. et al. 2007. Relating root structure and anatomy to whole-plant functioning in 14 herbaceous Mediterranean species. – New Phytol. 173: 313–321.
Iversen, C. M. et al. 2017. A global fine-root ecology database to address below-ground challenges in plant ecology. – New Phytol. 215: 15–26.
Kattge, J. et al. 2020. TRY plant trait database-enhanced coverage and open access. – Global Change Biol. 26: 119–188.
Kramer-Walter, K. R. et al. 2016. Root traits are multidimensional: specific root length is independent from root tissue density and the plant economic spectrum. – J. Ecol. 104: 1299–1310.
Kuchenbuch, R. et al. 2006. Effects of decreasing soil water content on seminal and lateral roots of young maize plants. – J. Plant Nutr. Soil Sci. 169: 841–848.
Laliberté, E. 2017. Below-ground frontiers in trait-based plant ecology. – New Phytol. 213: 1597–1603.
Laliberté, E. and Shipley, B. 2012. FD: measuring functional diversity from multiple traits, and other tools for functional ecology. – R package ver. 1.0-12. <http://CRAN.R-project.org/package=FD>.
Lecompte, F. et al. 2005. Patterns of variability in the diameter of lateral roots in the banana root system. – New Phytol. 167: 841–850.
Liu, Y. et al. 2018. A new method to optimize root order classification based on the diameter interval of fine root. – Sci. Rep. 8: 2960.
Ma, Z. et al. 2018. Evolutionary history resolves global organization of root functional traits. – Nature 555: 94–97.
McCormack, M. L. and Iversen, C. M. 2019. Physical and functional constraints on viable belowground acquisition strategies. – Front. Plant Sci. 10: 1215.
McCormack, M. L. et al. 2012. Predicting fine root lifespan from plant functional traits in temperate trees. – New Phytol. 195: 823–831.
McCormack, M. L. et al. 2015. Redefining fine roots improves understanding of below-ground contributions to terrestrial biosphere processes. – New Phytol. 207: 505–518.
Navas, M. L. et al. 2010. Suites of plant traits in species from different stages of a Mediterranean secondary succession. – Plant Biol. 12: 183–196.
Ottaviani, G. et al. 2020. The neglected belowground dimension of plant dominance. – Trends Ecol. Evol. 35: 763–766.
Pagliai, M. and De Nobili, M. 1993. Relationship between soil porosity, root development and soil enzyme activity in cultivated soils. – Geoderma 56: 243–256.
Pierce, S. et al. 2017. A global method for calculating plant CSR ecological strategies applied across biomes world-wide. – Funct. Ecol. 31: 444–457.
Poorter, H. et al. 2012. Biomass allocation to leaves, stems and roots: meta-analyses of interspecific variation and environmental control. – New Phytol. 193: 30–50.
Poorter, H. et al. 2015. How does biomass distribution change with size and differ among species? An analysis for 1200 plant species from five continents. – New Phytol. 208: 736–749.
Qin, R. et al. 2004. Impact of tillage system on winter wheat. – Agron. J. 96: 1523–1530.
Rasmann, S. et al. 2011. Predicting root defence against herbivores during succession. – Funct. Ecol. 25: 368–379.
Reich, P. B. 2014. The world-wide 'fast–slow' plant economics spectrum: a traits manifesto. – J. Ecol. 102: 275–301.
Roumet, C. et al. 2016. Root structure–function relationships in 74 species: evidence of a root economics spectrum related to carbon economy. – New Phytol. 210: 815–826.
Ryser, P. and Lambers, H. 1995. Root and leaf attributes accounting for the performance of fast-and slow-growing grasses at different nutrient supply. – Plant Soil 170: 251–265.
Ryser, P. et al. 2011. Constraints of root response to waterlogging in *Alisma triviale*. – Plant Soil 343: 247–260.
Scanlan, C. A. and Hinz, C. 2010. Using radius frequency distribution functions as a metric for quantifying root systems. – Plant Soil 332: 475–493.
Shao, X. et al. 2019. Future direction of searching for root economics spectrum: focusing on the fibrous roots 'absorptive unit'. – Ecosphere 10: e02716.
Soudzilovskaia, N. A. et al. 2020. FungalRoot: global online database of plant mycorrhizal associations. – New Phytol. 227: 955–966.
Tedersoo, L. et al. 2020. How mycorrhizal associations drive plant population and community biology. – Science 367: eaba1223.
Tilman, D. 1985. The resource-ratio hypothesis of plant succession. – Am. Nat. 125: 827–852.
Tjoelker, M. G. et al. 2005. Linking leaf and root trait syndromes among 39 grassland and savannah species. – New Phytol. 167: 493–508.
Valverde-Barrantes, O. J. et al. 2017. A worldview of root traits: the influence of ancestry, growth form, climate and mycorrhizal association on the functional trait variation of fine-root tissues in seed plants. – New Phytol. 215: 1562–1573.
Wahl, S. and Ryser. P. 2000. Root tissue structure is linked to ecological strategies of grasses. – New Phytol. 148: 459–471.
Wang, R. et al. 2018. Different phylogenetic and environmental controls of first-order root morphological and nutrient traits: evidence of multidimensional root traits. – Funct. Ecol. 32: 29–39.
Wardle, D. A. et al. 1998. Can comparative approaches based on plant ecophysiological traits predict the nature of biotic interactions and individual plant species effects in ecosystems. – J. Ecol. 86: 405–420.
Weemstra, M. et al. 2016. Towards a multidimensional root trait framework: a tree root review. – New Phytol. 211: 1159–1169.
Westoby, M. 1998. A leaf–height–seed (LHS) plant ecology strategy scheme. – Plant Soil 199: 213–227.
Zirbel, C. R. et al. 2017. Plant functional traits and environmental conditions shape community assembly and ecosystem functioning during restoration. – J. Appl. Ecol. 54: 1070–1079.
Zobel, R. W. 2013. *Lolium perenne* L. root systems are a collection of Gaussian curve shaped meso diameter class length distributions. – Plant Soil 363: 113–121.