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Ecosystems

ISSN 1432-9840

Ecosystems
DOI 10.1007/s10021-015-9946-x

Springer

Ecosystems
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ECOSYSTEMS

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Abstract

Climate change-induced rainfall reductions in Mediterranean forests negatively affect the decomposition of plant litter through decreased soil moisture. However, the indirect effects of reduced precipitation on litter decomposition through changes in litter quality and soil microbial communities are poorly studied. This is especially the case for fine root litter, which contributes importantly to forests plant biomass. Here we analyzed the effects of long-term (11 years) rainfall exclusion (29% reduction) on leaf and fine root litter quality, soil microbial biomass, and microbial community-level physiological profiles in a Mediterranean holm oak forest. Additionally, we reciprocally transplanted soils and litter among the control and reduced rainfall treatments in the laboratory, and analyzed litter decomposition and its responses to a simulated extreme drought event. The decreased soil microbial biomass and altered physiological profiles with reduced rainfall promoted lower fine root—but not leaf—litter decomposition. Both leaf and root litter, from the reduced rainfall treatment, decomposed faster than those from the control treatment. The impact of the extreme drought event on fine root litter decomposition was higher in soils from the control treatment compared to soils subjected to long-term rainfall exclusion. Our results suggest contrasting mechanisms driving drought indirect effects on above-(for example, changes in litter quality) and belowground (for example, shifts in soil microbial community) litter decomposition, even within a single tree species. Quantifying the contribution of these mechanisms relative to the direct soil moisture-effect is critical for an accurate integration of litter decomposition into ecosystem carbon dynamics in Mediterranean forests under climate change.

Key words: carbon cycle; climate change; drought; litter functional traits; Mediterranean forests; rainfall exclusion; resilience; soil decomposers.

Introduction

Mediterranean ecosystems are among the most impacted by ongoing climate change (Sala and others 2000; Giorgi 2006). Current climate models predict a potential decline in annual rainfall of 20–30% in the Mediterranean Basin (Somot and others 2008), and the frequency of short-term extreme

Received 28 January 2015; accepted 23 October 2015;

Electronic supplementary material: The online version of this article (doi:10.1007/s10021-015-9946-x) contains supplementary material, which is available to authorized users.

Authors' contributions PGP, SH, and IP designed the study and performed research. PGP and IP conducted the data analysis. JMO contributed new methods. PGP, IP, JMO, and SH wrote the manuscript.

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Published online: 21 December 2015

drought events could be multiplied by three during the twenty-first century (Sheffield and Wood 2008). Such alterations in the amount and distribution of rainfall will eventually reduce soil water availability (Limousin and others 2009), affecting processes that control the ecosystem carbon (C) balance (Harper and others 2005). Stronger reductions in plant C uptake through CO_2 fixation than in C loss through plant respiration processes under reduced rainfall have been hypothesized to trigger a positive C-climate feedback in Mediterranean (Misson and others 2010) and temperate forests (Ciais and others 2005). However, such estimates of ecosystem C balance based on ecosystem-level net fluxes do not allow a detailed evaluation of the relative importance of the different pathways of C fluxes, such as autotrophic (that is, plants) versus heterotrophic respiration (that is, soil decomposers). The latter contributes between 50 and 70% to total ecosystem respiration (Curiel Yuste and others 2005) and is fueled mostly by the microbial decomposition of plant-derived organic matter.

Rainfall exclusion experiments have typically found a negative effect on leaf litter decomposition as a consequence of a direct moisture-induced reduction in soil heterotrophic activity (Sanaullah and others 2012; Saura-Mas and others 2012; Walter and others 2013). However, the relative contribution of the longer term indirect consequences of reduced rainfall on litter quality (that is, the combination of chemical and morphological traits) and microbial decomposers (abundance, composition, physiology) is poorly understood, despite their importance in determining the overall outcome of climate change effects on litter decomposition (Allison and others 2013; Vogel and others 2013). In addition, soil microbial strategies relating to drying-rewetting cycles (that is, soil rewetting after a drought event) can shift from sensitive to tolerant after long-term exposure to rainfall reduction (Evans and Wallenstein 2014). Such microbial acclimation and/or adaptation can modify the resilience of microbially mediated processes to short-term extreme drought events (De Vries and others 2012). The importance of such modifications for litter decomposition, however, remains unclear, mostly because of a lack of long-term studies manipulating precipitation over a decade or more. These indirect effects through litter quality and microbial adjustments need to be addressed to establish a mechanistic understanding of the impacts of changing rainfall patterns on litter decomposition and ecosystem C dynamics in Mediterranean forests.

Although annual inputs of fine root litter can be equivalent to that of leaf litter (Jackson and others 1997; Freschet and others 2013)—especially in forests (Norby and others 2004)—the literature on litter decomposition is clearly biased towards leaves, with only 2% of all studies looking at roots (Zhang and others 2008). In Mediterranean evergreen forests, the need to fill this gap is especially urgent with ongoing climate change, as C allocation to fine roots has been found to increase with water stress (Canadell and Roda 1991). Although fine root litter seems to be more recalcitrant than leaf litter from the same plant species (Zhang and others 2008; Freschet and others 2012), the quality of leaf and fine root litter and their decomposition rates are often positively correlated (Wang and others 2010; Birouste and others 2012; Freschet and others 2012, but see Hobbie and others 2010), which may indicate similar and parallel responses to climate change. Rainfall reduction can decrease leaf litter quality in Mediterranean holm oak via increases in secondary metabolites (for example, phenolics and condensed tannins) and decreases in nutrient concentrations (Ogaya and Peñuelas 2006; Sardans and Peñuelas 2005), but whether such effects also occur for fine root litter is less well known. In addition, even if leaf and fine root litter quality responded similarly to reductions in rainfall, the consequences of such climate change-induced alterations in litter quality for microbial decomposers could differ between litter materials. Thus, due to the important contribution of fine roots to total plant biomass in forests, and the potential for contrasting responses compared to leaf litter, belowground plant organs should be taken into account when assessing the indirect role of litter quality and soil microbes in determining the impact of reduced rainfall on decomposition.

To disentangle the relative importance of changes in litter quality and differences in heterotrophic microbial communities for the response of litter decomposition to reduced rainfall, we took advantage of a long-term (11 years) rainfall exclusion experiment in a Mediterranean evergreen forest dominated by holm oak (*Quercus ilex* L.). Specifically, we evaluated the effects of reduced rainfall on above- and belowground litter traits, and on soil microbial biomass and community-level physiological profiles. We then conducted a reciprocal transplant experiment in the laboratory in which we placed leaf and fine root litter on their soil of origin (control or rainfall exclusion treatment) and on the soil of the contrasting treatment, and analyzed C mineralization as a measure of litter decomposition. At the end of a 95-day incubation period, all treat-

ment combinations were additionally exposed to a simulated extreme drought event to test for potential changes in the impacts on litter decomposition. We tested the following hypotheses: (i) long-term rainfall reduction decreases leaf and fine root litter quality by increasing their C:N ratios and the concentrations of secondary compounds, promoting a negative influence on litter decomposition, (ii) long-term rainfall reduction decreases soil microbial biomass, and alters microbial community-level physiological profiles, promoting a negative influence on litter decomposition, and iii) the impacts of extreme drought on litter decomposition are higher in soils not subjected to long-term rainfall reduction.

Materials and Methods

Study Area and Experimental Design of the Long-Term Rainfall Exclusion Experiment

The study area is located in an evergreen Mediterranean forest in southern France (3°35′45″E, 43°44′29″N at an elevation of 270 m). The vegetation and the litter layer are dominated by holm oak individuals and leaves, respectively. The climate is Mediterranean, with cool winters and a severe summer drought (MAT: 13.1°C, MAP: 901 mm). The soil (see Table 1 for detailed characteristics), dominated by Jurassic limestone, has a large volumetric rock content that reduces the total soil available water accumulated over 4.5-m depth to only 150 mm, and enhances the drought stress experienced by the vegetation during the summer (Rambal and others 2003). In 2003, a rainfall exclusion experiment was set up in the study area to simulate the effects of reductions in precipitation on Mediterranean evergreen forests. The experiment was replicated on three independent locations with at least 100 m between them. This design allowed us to account for the potential environmental heterogeneity in microclimate and soil conditions between locations. Each location was divided in two consecutive plots of 140 m^2 (14 m × 10 m), corresponding to the reduced rainfall (29% reduction of net precipitation achieved using PVC gutters covering a 33% area) and control (natural rainfall conditions, but identical gutters set up upside down) treatments. See Online Appendix S1 and Limousin and others (2009) for a detailed explanation of the rainfall exclusion facility.

Table 1. Soil Parameters of the Experimental Site

	Control	Drought
Total organic C (g kg^{-1})	86.0 ± 3.73	85.9 ± 4.79
Total N (g kg^{-1})	4.6 ± 0.24	4.7 ± 0.21
Olsen P (mg kg^{-1})	22.3 ± 1.68	23.8 ± 1.41
NO_3^-–N (mg kg^{-1})	4.4 ± 0.58	6.2 ± 1.18
NH_4^+–N (mg kg^{-1})	36.4 ± 2.90	34.7 ± 1.92

Mean ± 1 SE, n = 15.

Sampling of Leaf Litter, Fine Root Litter, and Soils

We sampled fresh fallen leaf litter, fine roots, and soils from specifically marked individual trees to tease apart the contribution of litter quality and soil microbes on litter decomposition responses to long-term reductions in rainfall amount accounting for intra-specific variability in leaf and fine root litter traits. The collected litter material was used to determine quality traits and to set up a laboratory reciprocal transplant experiment incubating litter (leaf and fine root) and soil from the two treatments (control and exclusion) of the rainfall exclusion field experiment. Five *Q. ilex* individual trees per plot were sampled for a total of n = 30 trees (two rainfall treatments × three locations × 5 individual trees). The five trees were selected at a minimum distance of 4–5 m to any other tree/shrub individuals to assure that soil beneath the canopy of sampled individual trees were little influenced by neighboring trees. Mesh litter traps were placed within the canopy of each sampled *Q. ilex* individual as high as possible in order to assure the collection of leaf litter originating from the particular target individual. These litter traps were placed in the field in early September 2013 and retrieved in late October 2013 to include the second litterfall peak of *Q. ilex*, which is likely to be more influenced by the reduced rainfall treatment than the spring peak, as the experimental rainfall reduction especially decreases soil moisture during summer drought (Limousin and others 2009). Intact leaves were selected and oven-dried for three days at 40°C for litter quality and decomposition measurements.

Three soil cores (5-cm diameter, 15-cm deep) were collected at three random positions beneath the canopy of each individual tree at the same time as we collected leaf litter from the litter traps. These soil samples were used for soil microbial analyses and fine root collection. Upon harvest, the three soil cores of each individual tree were mixed (30 samples in total), sieved at 2 mm, and separated into three subsamples. One subsample was immediately frozen at −20°C for microbial analyses, a second subsample was air-dried for measurements

of soil parameters, and the third one was stored at 4°C for three weeks until the decomposition experiment started. During soil sieving, we retrieved fine roots (diameter < 2 mm) that were gently washed to eliminate adhered soil particles. Once washed, *Q. ilex* roots were sorted visually from other species' roots (usually less than 5% of the total fine root pool), to prepare a homogeneous pool of fine roots for each individual tree. It is reasonable to assume that the fine root pool was dominated by this particular individual tree since neighboring trees were at least 4–5 m apart. However, we cannot entirely exclude that the fine root pool also contained some roots from neighboring *Q. ilex* trees. We did not attempt to distinguish between live and dead roots, as this is practically impossible for our study species, and most studies have reported little or no difference in nutrient content between them (McClaugherty and others 1982; Aerts 1990). Immediately after collection, roots were separated into three subsamples. Two subsamples were oven-dried at 40°C for 3 days and used for litter chemistry and the decomposition experiment, and the third subsample was immersed in tap water and stored at 4°C for root morphology measurements.

Leaf and Fine Root Litter Quality, Soil Microbes, and Soil Parameters

The oven-dried subsamples of leaf and fine root litter from each *Q. ilex* individual (60 samples in total) were ground to fine powder with a ball mill. C and nitrogen (N) concentrations were determined using a CN elemental analyzer (ThermoFinnigan, Milan, Italy). Total phenolics were measured with the Folin–Ciocalteu reagent following Marigo (1973), but using methanol (50%) as solvent instead of water. Condensed tannins were determined according to the acid butanol method (Porter and others 1986). Specific leaf area (SLA) and its belowground counterpart, specific root length (SRL), were measured as morphological traits. We measured SLA (leaf area/dry mass) by collecting leaf disks (0.24 cm^2) from five randomly selected leaves per tree individual (Pérez-Harguindeguy and others 2013). The five leaf disks were pooled, dried at 60°C for two days, and weighted (that is, one mean value per individual tree). This technique likely overestimated SLA because structural masses (for example, veins), which may increase leaf weights, were avoided across treatments. To measure SRL (total root length/dry mass), roots were spread out in distilled water onto a mesh tray, carefully dried to eliminate excess water and finally transferred on a transparent acetate sheet, and scanned at 400 dpi. The resulting image was processed with image analysis software (Winrhizo, version 2009, Regent Instrument, Quebec, Canada) to determine total root length (L). After scanning, roots were oven-dried at 60°C for 72 h and then weighed to determine their dry mass.

We analyzed the functional composition of soil heterotrophic microbial communities with the MicroResp system, which is a whole-soil method based on community-level physiological profiles obtained by testing ecologically meaningful carbon sources of different chemical recalcitrance (Campbell and others 2003). In functional terms, the substrate utilization rates of the C sources correspond to the catabolic attributes of the microbial community, and thus we can use MicroResp data to interpret differences in microbial functional composition (García-Palacios and others 2011a). Before MicroResp measurements, previously frozen (−20°C) soil was incubated in 96-DeepWell Microplates for 5 days at 25°C and at 50% of their water-holding capacity in order to allow microbial communities to reestablish in defrosting soil. The appropriateness of storing the soil samples frozen for measurements of microbial activity has been demonstrated with substrate-induced respiration techniques (Pesaro and others 2003), a procedure very similar to the MicroResp. We then followed the procedure described in García-Palacios and others (2011b) to calculate substrate-induced respiration rates expressed in μg C–CO_2 respired g^{-1} soil h^{-1} by using the control (deionized water but no C source added) as the basal respiration. Fifteen different C sources were added: three carbohydrates (D-glucose, xylan, cellulose), one amine (*N*-acetyl-glucosamine), five amino acids (L-asparagine, L-glutamine, L-lysine, L-serine, L-glycine), three carboxylic acids (malic acid, oxalic acid, uric acid), and three phenolic acids (caffeic, syringic and vanillic). Active microbial biomass was estimated by converting glucose-induced respiration rates obtained with the MicroResp system to biomass. For soil parameters, soil subsamples were sent to the INRA laboratory at Arras, France, for standard soil analyses (pH, total C, total N, Olsen P, NH_4^+–N and NO_3^-–N).

Leaf and Fine Root Litter Decomposition Assay

Using the leaf litter, fine root litter, and soil samples collected in the field, we set up a reciprocal transplant experiment in the laboratory, with the five individual trees sampled per plot as replicates (nested within locations). We evaluated whether litter origin (for example, changes in chemistry

and/or morphology between control and rainfall exclusion) and soil origin (for example, changes in microbial abundance and/or composition between control and rainfall exclusion) mediated the responses of leaf and fine root litter decomposition to reduced rainfall. These main effects were crossed in a fully factorial design and replicated for the five trees within each location of the field experiment (two litter origins × two soil origins × three locations × 5 individual trees, $n = 60$ for each litter material). Thus, individual tree-level samples of leaf litter or fine root litter from each litter origin were incubated with either the same (control litter on control soil, rainfall exclusion litter on rainfall exclusion soil) or different soil origin (control litter on rainfall exclusion soil, rainfall exclusion litter on control soil). The 'same origin' treatment consisted of litter and soil from the same individual tree, whereas in the 'different origin' treatment, we allocated tree-level litter randomly to soil from an individual tree from the other treatment, but within the same location. The comparison of soil origins within each pair of control and rainfall exclusion plots kept environmental heterogeneity (microclimate, soil physicochemical parameters), and therefore unaccounted variance, to a minimum, increasing our ability to assign soil origin effects to differences in soil microbes. To minimize potential non-microbial soil effects on decomposition, such as soil fertility, we used a higher litter to soil ratio (0.75 g:20 g) than previous experiments (Ayres and others 2009; García-Palacios and others 2013) for the construction of the microcosms (150-ml flasks). For standardization, both leaf and fine root litter were placed on top of the soil surface in each microcosm assuring a good contact with the soil surface. We regularly measured C mineralization rates, a measure of litter decomposition over the incubation period of a total of 95 days using a Micro-Gas Chromatograph (Varian GC 4900, Walnut Creek, USA). One 'no-litter' (that is, containing only soil) microcosm per individual tree ($n = 30$) was also incubated to correct for the soil contribution to CO_2 production. We used linear interpolations between sampling dates and then summed them across all dates to estimate the cumulative CO_2 produced (C loss) over the incubation period.

Impacts of an Extreme Drought Event on Leaf and Fine Root Litter Decomposition

A week after the last respiration measurements on day 95, we exposed all microcosms to a simulated extreme drought event, in order to assess whether the impact of such an event on litter decomposition was influenced by litter and soil origins from the long-term precipitation exclusion experiment. We simulated the extreme drought event at the end of the 95-day incubation period for two reasons: (i) to enable a direct comparison of pre- and post-drought measurements at the level of each individual microcosm (avoiding treatment unrelated variation among microcosms), and (ii) to reduce the magnitude of the conspicuous respiration flush that is commonly found in soils subjected to air-drying and rewetting (the Birch effect; Birch 1958). The duration and intensity of this drought event was determined based on the most extreme drought recorded at the study site (July–August 2006, Limousin and others 2012). Microcosms were kept open in the growth chamber at 20°C during 24 days to a final water-holding capacity (WHC) of 5–8%. Successively, all microcosms were rewetted to 50% WHC, and C mineralization rates were measured at 1, 3, 7, and 15 days after rewetting. We used the C mineralization rate measured at day 95, the date immediately before the extreme drought event was initiated, as a reference to calculate a Drought Impact Index (DII hereafter) for each of the four sampling dates. The DII for each microcosm was calculated as the increase in C mineralization rate after the extreme drought event (1, 3, 7, or 15 days) relative to the pre-drought measurements on day 95. We would expect the DII to be equal to or higher than 1 because of the CO_2 flush from soils after rewetting, which is thought to be higher with a more negative impact of the drought event (Fierer and Schimel 2003). This index allowed us to compare the impact of extreme drought events on litter decomposition as a function of the soil and litter origin treatments, by standardizing the potential differences in process rates between treatments and focusing on the rate of change in response to the applied disturbance.

Data Analysis

To reduce the number of tests conducted, and allow for an integrative interpretation of the effects of the long-term field rainfall exclusion treatment on leaf and fine root litter quality, we built two separate trait matrices, one for leaf litter (total phenolics, condensed tannins, C:N ratio and SLA) and one for fine root litter (total phenolics, condensed tannins, C:N ratio and SRL). The effects of the rainfall exclusion on litter quality were evaluated using semiparametric permutational ANOVA-type tests (PERMANOVA, Anderson 2001). We used the Euclidean distance, 9999 permutations, *rainfall exclusion* as a fixed factor, and two random effects

(*location* and *tree* nested within *location*). This design allowed us to account for non-independence of individual trees within locations. Separate analyses were conducted for each litter material. The same PERMANOVA model was used to test for effects of rainfall exclusion on soil microbial community-level physiological profiles. To help visualize the effects of rainfall exclusion on the multivariate litter quality matrices, we also performed a principal coordinate analysis on these variables (PCO; Anderson and others 2008). We analyzed C mineralization rates using two (one for leaf and one for fine root litter) general linear mixed-effects models (GLMMs). Both models included *litter origin*, *soil origin*, and *sampling date* as fixed factors (along with their interactions), and three random effects (*location*, *tree* nested within *location*, and *microcosm* nested within *tree* nested within *location*). This design allowed us to account for non-independence of individual trees within locations (as litter transplants were performed at the level of the location, that is, two paired plots) and across sampling dates (as each microcosm was measured once on each date). The effects of varying litter origin and soil origin on cumulative C loss over the course of the 95-day decomposition experiment were assessed with similar GLMMs (but without *sampling date* and *microcosm)*. The same GLMMs used for C mineralization rates were also used to evaluate the effects of litter and soil origin on the DII. When interactions including the number of days after rewetting (*sampling date*) were significant, we ran a model for each date separately. Relationships between leaf and fine root litter traits, and between leaf and fine root litter cumulative C loss across treatments were analyzed with Pearson correlations. PERMANOVA and PCO analyses were carried out using the PERMANOVA + module for the PRIMER software (PRIMER-E Limited, Plymouth Marine Laboratory, UK (Anderson and others 2008)), and GLMMs were conducted with the R software environment (R Development Core Team 2011).

Results

Effects of Rainfall Exclusion on Soil Microbes and Traits of Leaf and Fine Root Litter

The rainfall exclusion (29% reduction of annual net precipitation) over 11 years had a marginally significant effect on the soil microbial community physiological profile ($F_{1,2}$ = 3.97, P = 0.051); this was related to lower respiration rates of the C sources syringic acid, vanillic acid, glycine, asparagine, and glutamine found in soils from rainfall exclusion plots (Figure 1A). The active microbial biomass was 27% lower in the soils from rainfall exclusion plots ($F_{1,2}$ = 28.23, P = 0.034; Figure 1B). The effect of reduced rainfall on leaf litter quality was marginally significant ($F_{1,2}$ = 5.09, P = 0.055), with leaf litter from trees in rainfall

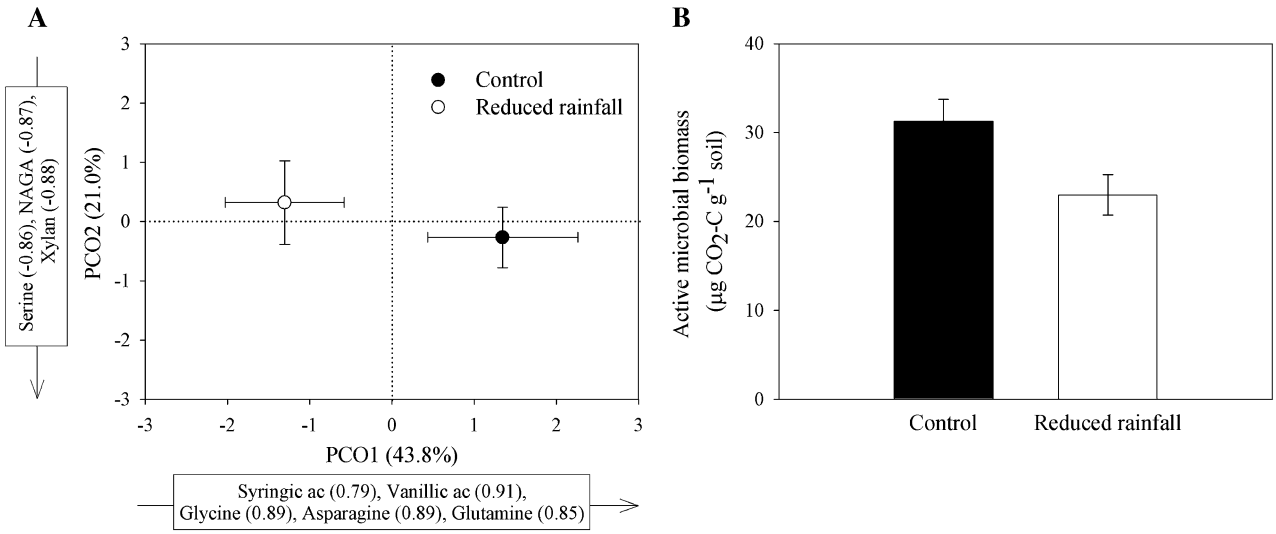

Figure 1. Effects of long-term rainfall exclusion in the field on **A** soil microbial community-level physiological profiles and **B** active microbial biomass (glucose-induced respiration rates). Variance explained by each ordination axes (PCO1 and PCO2) is shown in **A**, and significant Pearson correlations ($r \geq 0.7$) between individual substrates and the ordination axes are shown in the boxes, with the arrow representing the sign of the correlation. Values represent means ± 1 SE (n = 15).

exclusion plots showing lower concentrations of total phenolics and condensed tannins (Figure 2A; Table 2). Fine root litter quality was not significantly influenced by the reduced rainfall ($F_{1,2}$ = 0.55, P = 0.644; Figure 2B; Table 2). We found no significant correlations between leaf and fine root litter traits across the rainfall exclusion treatments ($P > 0.05$ in all cases, except for a positive correlation between SLA and SRL; r = 0.51, P = 0.004, Table S1 in Online Appendix).

Effects of Rainfall Exclusion on Leaf and Fine Root Litter Decomposition

Although the litter origin × soil origin interaction was not significant for either leaf or fine root litter decomposition (Table S2 in Online Appendix), we found differences in the main effects for both litter materials. Litter origin, but not soil origin, had a significant effect (P = 0.041, Table S2 in Online Appendix) on leaf litter C mineralization rates. This effect, however, depended on the incubation stage, as leaf litter from the rainfall exclusion treatment decomposed faster than the control litter only at the intermediate stages (litter origin × sampling date, P = 0.018, Figure 3B). This incubation time-dependent effect, nevertheless, led to a 31% increase in the cumulative C loss in the microcosms containing leaf litter from the reduced rainfall treatments (P = 0.003, Table S3 in Online Appendix). Contrary to the leaf litter, fine root litter C mineralization rates differed with the origin of the soil ($P < 0.001$, Table S2 in Online Appendix). Mineralization rates were lower in soils taken from

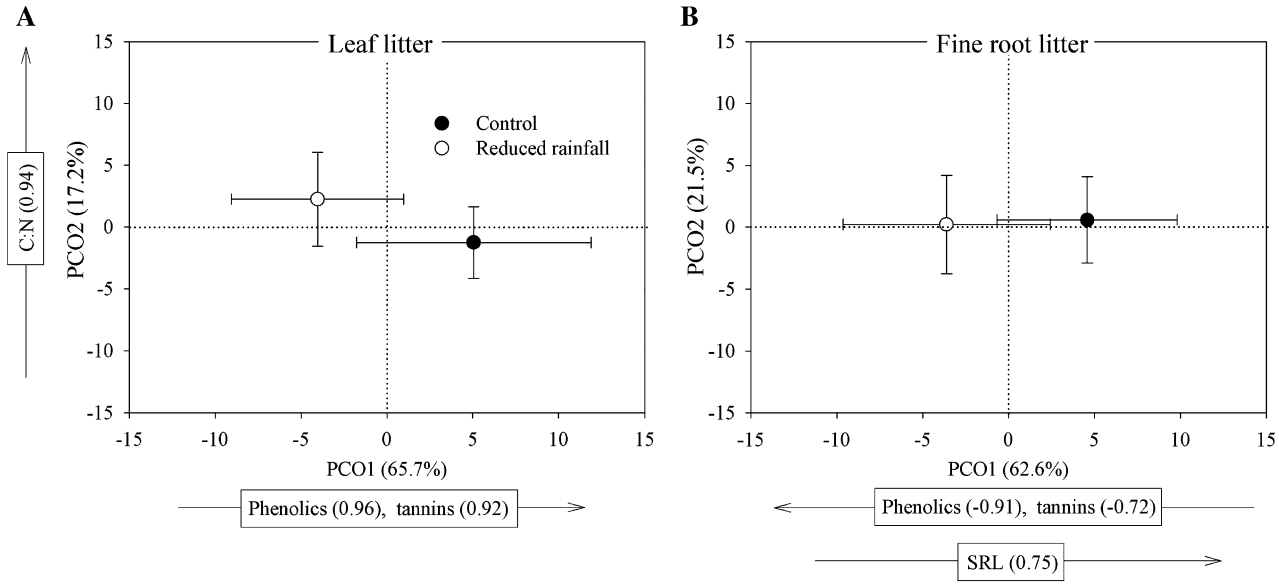

Figure 2. Effects of long-term rainfall exclusion in the field on **A** leaf and **B** fine root litter traits. Variance explained by each ordination axes (PCO1 and PCO2) is shown. Significant Pearson correlations ($r \geq 0.7$) between the original traits and the ordination axes are also shown in the boxes, with the arrow representing the sign of the correlation. Values represent means ± 1 SE (n = 15).

Table 2. Litter Functional Traits of *Quercus ilex* Individuals

Litter material	Rainfall exclusion	Total phenolics (% dry mass)	Condensed tannins (% dry mass)	C:N ratio	SLA ($mm^2\ mg^{-1}$) or SRL ($m\ g^{-1}$)
Leaf	Control	5.7 ± 0.56	9.4 ± 1.01	57.0 ± 2.16	3.9 ± 0.15
	Reduced rainfall	5.0 ± 0.45	8.1 ± 0.41	60.6 ± 2.77	3.8 ± 0.11
Fine root	Control	9.7 ± 0.60	20.2 ± 0.75	76.8 ± 3.42	4.5 ± 0.51
	Reduced rainfall	10.0 ± 0.61	23.9 ± 2.19	78.2 ± 2.47	3.8 ± 0.31

Mean ± 1 SE, n = 15.

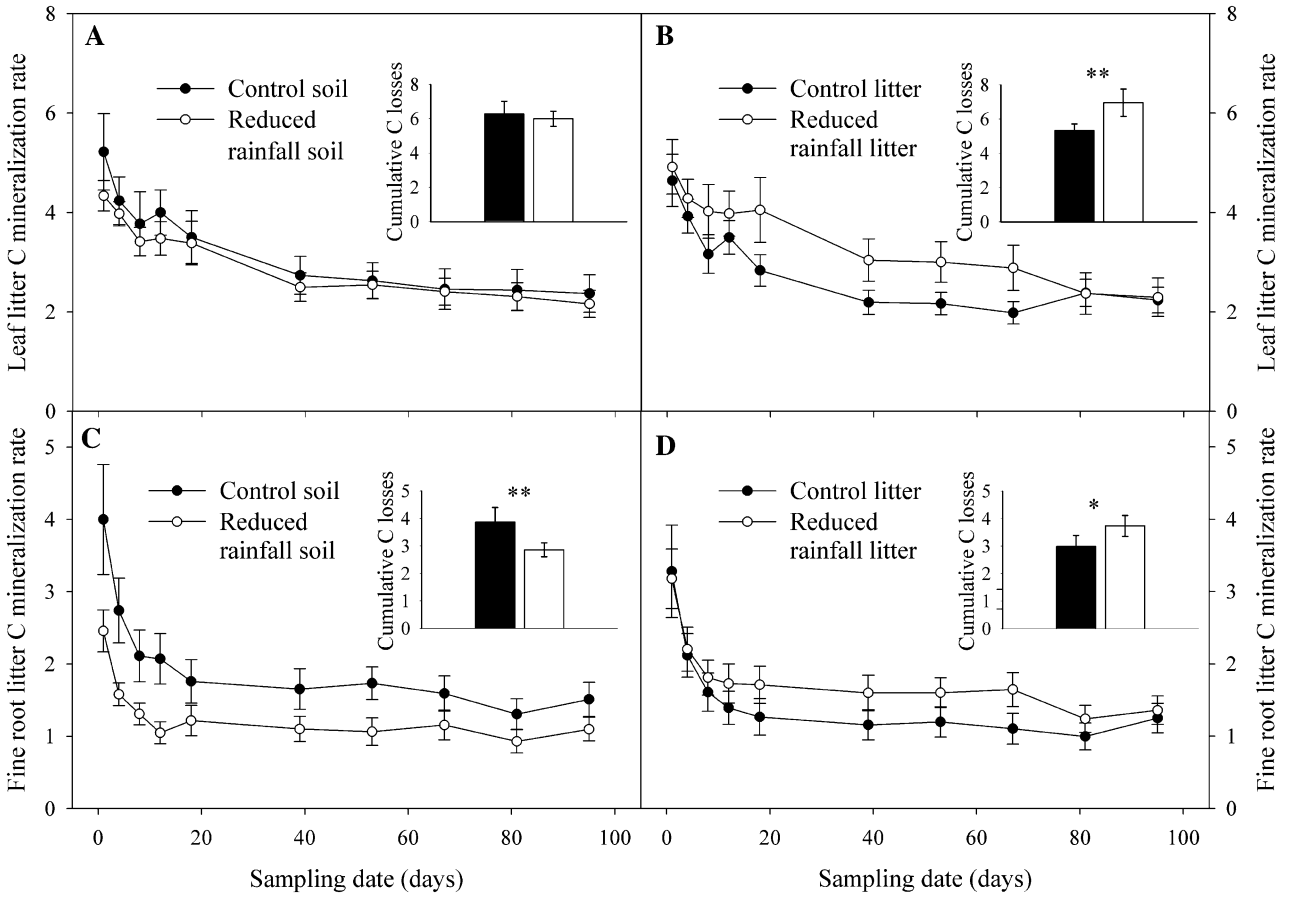

Figure 3. Leaf litter C mineralization rates (μg CO_2–C g^{-1} soil h^{-1}) in response to long-term rainfall manipulation for **A** the soil origin and **B** the litter origin. Fine root litter C mineralization rates in response to long-term rainfall manipulation for **C** the soil origin and **D** the litter origin. The effects of soil and litter origins represent the effects of changes in soil microbes and litter quality, respectively, between rainfall exclusion levels, and were analyzed separately between leaves and fine roots. Mean values (±1 SE) across litter origins are shown in **A** and **C**, and mean values across soil origins are shown in B and D for simplicity (n = 30). Inset bar charts represent the cumulative C losses (mg CO_2–C g^{-1} soil) over the course of the 95-day incubation experiment in response to long-term rainfall exclusion. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

the rainfall exclusion treatment, although this effect was only significant during the first 67 days of incubation (soil origin × sampling date, $P < 0.001$, Figure 3C). These differences resulted in a 26% lower cumulative C loss in the microcosms with soils from the rainfall exclusion treatment ($P = 0.008$, Table S3 in Online Appendix). Litter origin had no significant effect on C mineralization rates but there was a trend of higher rates with fine root litter originating from the rainfall exclusion treatment during most of the incubation period ($P = 0.099$, Table S2 in Online Appendix, Figure 3D). Over the entire period of incubation, these slightly higher C mineralization rates accumulated to a significant 25% higher C loss ($P = 0.044$, Table S3 in Online Appendix) for fine root litter from the rainfall exclusion treatment. The leaf and fine root litter cumulative C losses were positively correlated (Figure 4).

Impact of Extreme Drought on Litter Decomposition

Overall, the DII of leaf and fine root litter C mineralization rates were close to or higher than one over the entire measurement period and across treatments (Figure 5). Despite this general pattern, leaf litter C mineralization showed a higher DII for leaf litter material originating from the rainfall exclusion plots ($P = 0.041$, Table S4 in Online Appendix) at all the dates evaluated (Figure 5B). In contrast, there was no difference between fine root

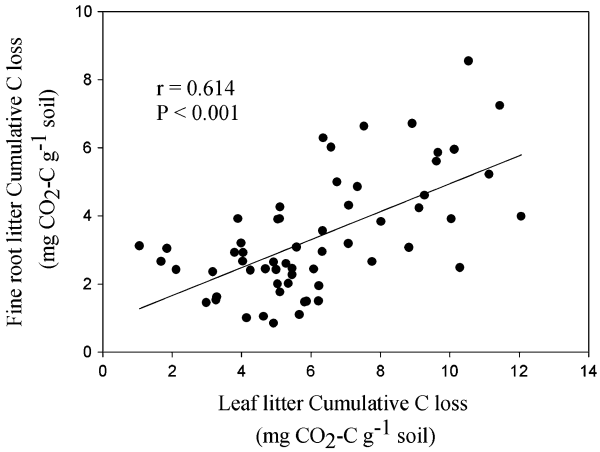

Figure 4. Across treatments Pearson correlation between the cumulative C loss from leaf and fine root litter over the course of the 95-day decomposition experiment (n = 60).

litters from the two origins (Figure 5D). Soil origin had no significant effect on the DII of leaf litter C mineralization rate, although there was a trend of higher impact indices in soils from control plots (Figure 5A). A similar pattern was observed for fine root litter C mineralization rates (Figure 5C). However, this difference decreased over time after the rewetting (soil origin × sampling date, $P < 0.001$, Table S4 in Online Appendix). The influence of rainfall exclusion was significant one day ($F_{1,42} = 4.19$, $P = 0.047$) and marginally significant three days ($F_{1,42} = 3.28$, $P = 0.077$) after rewetting, and ceased after that.

Discussion

Litter decomposition is generally negatively affected by diminished precipitation because of re-

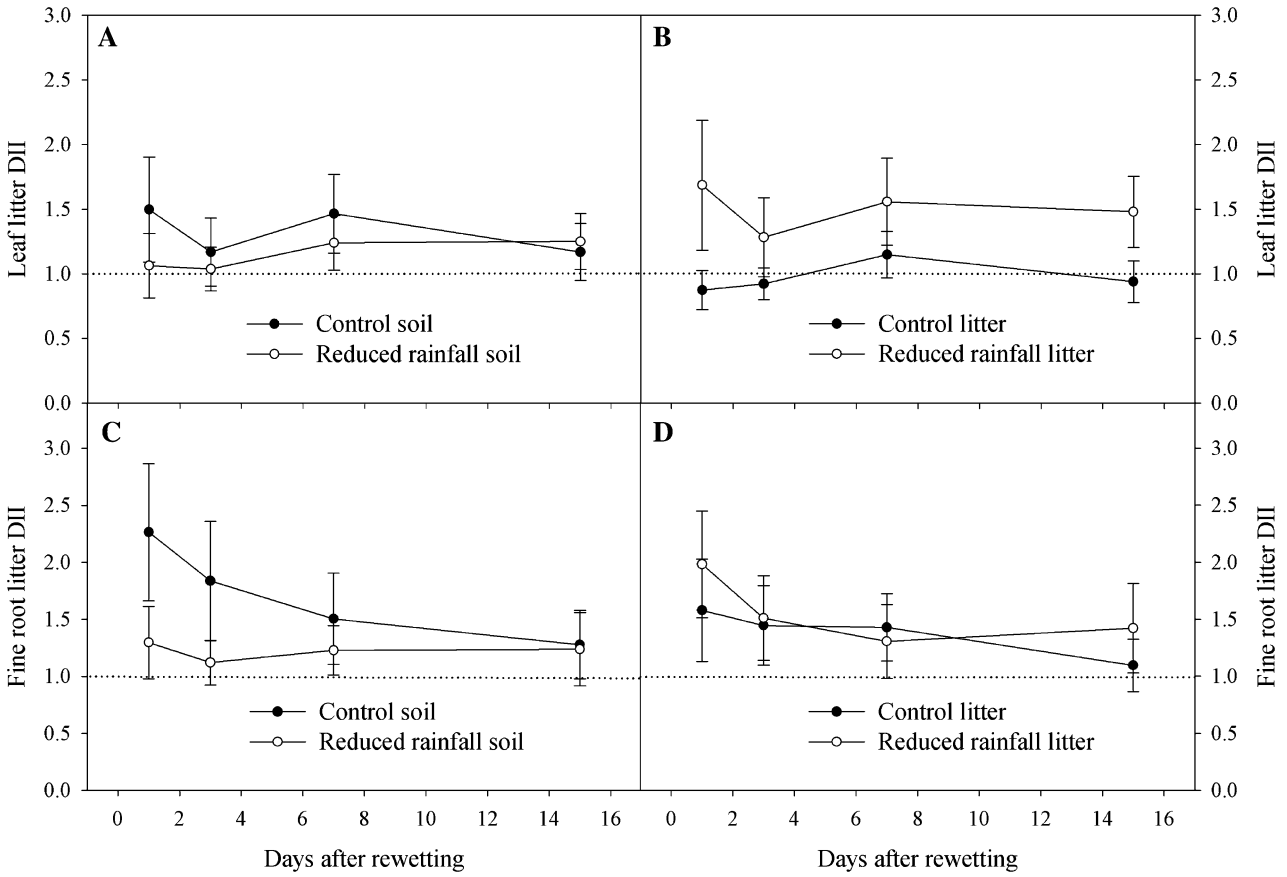

Figure 5. Drought Impact Index (DII, unitless) of leaf litter C mineralization rates in response to a laboratory-based extreme drought event for **A** the soil origin and **B** the litter origin. Drought Impact Index of fine root litter C mineralization rates in response to long-term rainfall manipulation for **C** the soil origin and **D** the litter origin. The effects of soil and litter origins represent the effects of changes in soil microbes and litter quality, respectively, between rainfall exclusion levels, and were analyzed separately between leaves and fine roots. DII = C mineralization rate 1, 3, 7, or 15 days after rewetting/C mineralization rate before extreme drought (day 95). Mean values (±1 SE) across litter origins are shown in **A** and **C**, and mean values across soil origins are shown in **B** and **D** for simplicity (n = 30).

duced soil moisture and therefore lower soil heterotrophic activity (Sanaullah and others 2012; Vogel and others 2013; Walter and others 2013). However, the importance of indirect drought effects via changes in litter quality and soil microbes is less well understood. Here we used a reciprocal transplant approach to distinguish the relative contribution of changes in soil microbial communities and litter quality on litter decomposition responses to reduced rainfall (~29% decrease in annual net precipitation) over more than 10 years in a Mediterranean evergreen forest. Our results indicate that both leaf and fine root litter decomposition are affected by these indirect effects in response to long-term rainfall reduction. However, the mechanisms driving these responses differed between above- and belowground plant organs. Our data suggest that reduced rainfall influences leaf litter decomposition mostly by changing the litter quality, while fine root litter decomposition is influenced by changes in the soil microbial community that developed in the rainfall exclusion plots that differed from the community developed in control plots.

The decreases in the amount of rainfall predicted with climate change can reduce leaf nutrient concentrations in Mediterranean holm oak, mostly through limited plant nutrient uptake as a result of lower soil mineralization rates (Sardans and Peñuelas 2005). Reduced rainfall can also increase the concentration of plant secondary metabolites (for example, phenolics and condensed tannins), as a consequence of a larger reduction in plant growth relative to photosynthesis (Ogaya and Peñuelas 2006). Such changes in tissue quality (lower nutrient concentrations and more secondary metabolites) can have cascading effects on ecosystem processes via their negative effect on microbial activity, reducing litter decomposition rates in Mediterranean forests exposed to climate change-induced drought (Sardans and Peñuelas 2007; Saura-Mas and others 2012). However, in contrast to our first hypothesis and to the results reported by Ogaya and Peñuelas (2006) for the same tree species, we observed slightly decreased concentrations of condensed tannins and phenolics in the leaf litter produced by trees subjected to long-term rainfall reductions. In contrast to leaf litter, we measured similar concentrations of phenolics and condensed tannins, and also similar C:N ratios in fine root litter of trees grown in control and rainfall exclusion plots, suggesting little impact of rain exclusion on fine root litter quality.

In line with our second hypothesis, soil microbial biomass was lower in soils sampled from the rainfall exclusion plots, and community-level physiological profiles shifted towards a decreased ability to catabolize both recalcitrant (for example, syringic and vanillic acids) and more labile C sources (for example, glycine, asparagine, and glutamine). A previous study in a similar holm oak forest in northern Spain reported lower sensitivity of soil fungal than bacterial communities to long-term rainfall reduction (Curiel Yuste and others 2011). These results suggest that the alteration in soil microbial functional composition found in our study may be largely determined by the low ability of bacterial communities to cope with drought in Mediterranean evergreen forests. A former study at the same experimental site where we took our samples documented a change in the composition of ectomycorrhizal fungal communities in response to reduced rainfall (Richard and others 2011). Changes in ectomycorrhizal communities are likely a direct result of decreasing water availability, representing an adaptation of the tree-mycorrhiza system to increasing drought that is critical for the trees' water balance (Kipfer and others 2012). These changes in ectomycorrhizal community structure are unlikely to have affected the results in our litter decomposition experiment, because alive plant roots, and therefore ectomycorrhizae, were excluded from the microcosms we used in the laboratory incubation.

The soil microbial responses to reduced rainfall reported here are consistent with the decrease in fine root litter C mineralization rates and cumulative C loss in soils from rainfall exclusion plots measured in our laboratory incubations. However, leaf litter decomposition was not affected by soil origin. The concentrations of secondary compounds may offer a possible explanation for these tissue-specific differences. The concentrations of total phenolics and condensed tannins in fine root litter were twice as high compared to those measured in leaf litter, irrespective of the precipitation treatment (Table 2). Such large differences in overall litter recalcitrance may suggest that the decomposition of the secondary compounds-rich root litter was more negatively affected by the reduced microbial ability to catabolize recalcitrant C sources involved in the degradation of polyphenols (for example, syringic and vanillic acids) under rainfall exclusion, than that of the more labile leaf litter. Moreover, leaf litter produced in rainfall exclusion plots showed reduced concentrations of secondary compounds which might have compensated for the reduced microbial capacity to break down secondary compounds consistent with the null effect of soil origin on leaf litter decomposition.

The reported effect of soil origin on fine root litter decomposition was not modified by the origin of fine root litter (there was no soil origin × litter origin interaction), indicating that soil microbes did not specialize on the decomposition of locally produced fine root litter. This result may be not so surprising since there were no apparent differences in fine root litter quality traits between control and rainfall exclusion plots, which consequently may not require any particular microbial adaptation for specific litter substrates (Allison and others 2013). However, like leaf litter, the fine root litter material originating in the rainfall exclusion plots also decomposed faster than that originating in the control plots (Figure 3), indicating that some quality characteristics we did not measure here, such as labile C compounds, lignin or calcium, differed between the two treatments. Interestingly, the 25% increase in cumulative C loss from fine root litter material collected in the rainfall exclusion plots was of similar magnitude to the 26% decrease in cumulative C loss when fine root litter decomposed on soil from the rainfall exclusion treatment. Thus, the positive and negative effects of long-term rainfall exclusion on fine root litter cumulative C loss mediated by litter quality and soil microbes, respectively, canceled each other out.

The cumulative losses of C from leaf and fine root litter during the laboratory incubation were positively correlated across treatments, suggesting a strong relationship between the decomposition of above- and belowground tissues of the same tree individuals at a local scale. Similar positive relationships between leaf and fine root litter decomposition rates were previously suggested to reflect site-specific micro-climatic conditions (Wang and others 2010). However, in our experiment, soil moisture and temperature were kept constant among microcosms during the incubation period, suggesting that the correlation between fine root and leaf litter decomposition was instead driven by litter inherent characteristics, that is, co-varying litter traits. Previous studies conducted with herbaceous and woody species observed a positive correlation between leaf and root litter decomposition rates when the key traits determining decomposition covaried among above- and belowground plant organs (Wang and others 2010; Birouste and others 2012; Freschet and others 2012, but see Hobbie and others 2010). Despite correlating positively with fine root litter decomposition, the cumulative C loss from leaf litter was an order of magnitude greater than that from fine root litter from individuals of the same population of *Q. ilex* (Figures 3, 4). This variation between above- and belowground plant organs is similar to that found in multispecies experiments using mass loss as a measure of litter decomposition (Hobbie and others 2010; Freschet and others 2012). The intra-specific variation in decomposition between plant organs in monodominant forests such as the *Q.ilex* forest studied here, may be as important for certain ecosystem processes (for example, soil carbon and nitrogen cycling) as inter-specific differences in species-rich ecosystems (Coq and others 2010; Aerts and others 2012). These results support the emerging view that within-species variation in plant functional traits should be taken into account when predicting plant community and ecosystem functioning responses to global change (Violle and others 2012).

Across litter and soil origins, the Drought Impact Index (DII) showed values higher or close to one, suggesting the presence of Birch effects (that is, soil respiration flush) boosting initial CO_2 flushes after air-drying and rewetting (Xiang and others 2008). The Birch effect has been proposed to be driven by improved microbial access to physically protected organic matter (Denef and others 2001), and/or by increasing C substrate availability originating from dead microbial cells as a result of water potential stress (Fierer and Schimel 2003). When evaluating the influence of soil origin on the DII, however, it is unlikely that organic matter availability differs between control and reduced rainfall soils as both experienced the same air-drying and rewetting cycles. Thus, a higher DII likely represents a higher Birch effect due to increased breakdown of microbial cells from stress-sensitive microbes (Halverson and others 2000). The impact of extreme drought on leaf and fine root litter C mineralization rates, although only significant for the latter, was higher in control than in rainfall exclusion soils, suggesting that soil microbial communities from control plots were less resilient to extreme drought than those from the rainfall exclusion plots. These results suggest that microbes from the long-term rainfall exclusion plots may be more adapted to drought conditions, in line with the proposed selection for microorganisms capable to cope with drying-rewetting (for example, drought tolerance) by historical exposure to reduced rainfall (Evans and Wallenstein 2014).

Conclusions

We showed here, that in addition to direct moisture-driven effects on litter decomposition, climate change can also have indirect effects through alterations in the structure and functioning of soil microbial communities and in litter quality. In the studied holm oak coppice, the dominant forest type

among evergreen forests of the Mediterranean Basin (Terradas 1999), we found contrasting mechanisms driving indirect effects of reduced rainfall on above-(litter quality) and belowground (soil microbes) litter decomposition, even within the same tree species. Whether such indirect drought effects are relevant for ecosystem C dynamics in Mediterranean forests is presently unknown, and would require more detailed long-term measurements on C fluxes. Nevertheless, our results highlight the importance of accounting for climate change-induced indirect consequences on ecosystem processes beyond the obvious change in environmental conditions for an accurate prediction of climate-C cycle feedbacks.

ACKNOWLEDGEMENTS

We thank Noelia Portillo and Patrick Schevin for laboratory support. We would like to thank E.J. Sayer and an anonymous reviewer for their valuable comments and suggestions to improve the quality of this manuscript. All chemical and litter decomposition analyses (except the soil analyses specified in the text) were performed at the Plate-Forme d'Analyses Chimiques en Ecologie, LabEx Centre Méditerranéen de l'Environnement et de la Biodiversité. PGP was funded by the People Programme (Marie Curie Actions) of the European Union's Seventh Framework Programme (FP7/2007-2013) under REA Grant Agreement No DECOMFORECO-2011-299214.

REFERENCES

Aerts R. 1990. Nutrient use efficiency in evergreen and deciduous species from heathlands. Oecologia 84:391–7.

Aerts R, van Bodegom PM, Cornelissen JHC. 2012. Litter stoichiometric traits of plant species of high-latitude ecosystems show high responsiveness to global change without causing strong variation in litter decomposition. New Phytol 96:181–8.

Allison SD, Lu Y, Weihe C, Goulden ML, Martiny AC, Treseder KK, Martiny JBH. 2013. Microbial abundance and composition influence litter decomposition response to environmental change. Ecology 94:714–25.

Anderson MJ. 2001. A new method for non-parametric multivariate analysis of variance. Aust Ecol 26:32–46.

Anderson MJ, Gorley RN, Clarke KR. 2008. PERMANOVA+ for PRIMER: Guide to software and statistical methods. Plymouth: PRIMER-E.

Ayres E, Steltzer H, Berg S, Wall DH. 2009. Soil biota accelerate decomposition in high-elevation forests by specializing in the breakdown of litter produced by the plant species above them. J Ecol 97:901–12.

Birch HF. 1958. The effect of soil drying on humus decomposition and nitrogen. Plant Soil 10:9–31.

Birouste M, Kazakou E, Blanchard A, Roumet C. 2012. Plant traits and decomposition: are the relationships for roots comparable to those for leaves? Ann Bot 109:463–72.

Campbell CD, Chapman SJ, Cameron CM, Davidson MS, Potts JM. 2003. A rapid microtiter plate method to measure carbon dioxide evolved from carbon substrate amendments so as to determine the physiological profiles of soil microbial communities by using whole soil. Appl Environ Microbiol 69:3593–9.

Canadell J, Rodà F. 1991. Root biomass of *Quercus ilex* in a montane Mediterranean forest. Can J For Res 21:1771–8.

Ciais P, Reichstein M, Viovy N, Granier A, Ogee J, Allard V, Aubinet M, Buchmann N, Bernhofer C, Carrara A, Chevallier F, De Noblet N, Friend AD, Friedlingstein P, Grunwald T, Heinesch B, Keronen P, Knohl A, Krinner G, Loustau D, Manca G, Matteucci G, Miglietta F, Ourcival JM, Papale D, Pilegaard K, Rambal S, Seufert G, Soussana JF, Sanz MJ, Schulze ED, Vesala T, Valentini R. 2005. Europe-wide reduction in primary productivity caused by the heat and drought in 2003. Nature 437:529–33.

Coq S, Souquet JM, Meudec E, Cheynier V, Hättenschwiler S. 2010. Interspecific variation in leaf litter tannins drives decomposition in a tropical rain forest of French Guiana. Ecology 91:2088–91.

Curiel Yuste J, Nagy M, Janssens IA, Carrara A, Ceulemans R. 2005. Soil respiration in a mixed temperate forest and its contribution to total ecosystem respiration. Tree Physiol 25:609–19.

Curiel Yuste J, Peñuelas J, Estiarte M, Garcia-Mas J, Mattana S, Ogaya R, Pujol M, Sardans J. 2011. Drought-resistant fungi control soil organic matter decomposition and its response to temperature. Glob Chang Biol 17:1475–86.

De Vries F, Liiri M, Bjørnlund L, Bowker M, Christensen S, Setälä H, Bardgett R. 2012. Land use alters the resistance and resilience of soil food webs to drought. Nat Clim Chang 2:276–80.

Denef K, Six J, Paustian K, Merckx R. 2001. Importance of macroaggregate dynamics in controlling soil carbon stabilization: short-term effects of physical disturbance induced by dry–wet cycles. Soil Biol Biochem 33:2145–53.

Evans SE, Wallenstein MD. 2014. Climate change alters the ecological strategies of soil bacteria. Ecol Lett 17:155–64.

Fierer N, Schimel JP. 2003. A proposed mechanism for the pulse in CO_2 production commonly observed following the rapid rewetting of a dry soil. Soil Sci Soc Am J 67:798–805.

Freschet GT, Aerts R, Cornelissen JHC. 2012. A plant economics spectrum of litter decomposition. Funct Ecol 26:56–65.

Freschet GT, Cornwell WK, Wardle DA, Elumeeva TG, Liu W, Jackson BG, Onipchenko VG, Soudzilovskaia NA, Tao J, Cornelissen JHC. 2013. Linking litter decomposition of above- and below-ground organs to plant-soil feedbacks worldwide. J Ecol 101:943–52.

García-Palacios P, Bowker MA, Maestre FT, Soliveres S, Valladares F, Papadopoulos J, Escudero A. 2011a. Ecosystem development in roadside grasslands: biotic control, plant–soil interactions and dispersal limitations. Ecol Appl 21:2806–21.

García-Palacios P, Bowker MA, Chapman SJ, Maestre FT, Soliveres S, Gallardo A, Valladares F, Guerrero C, Escudero A. 2011b. Early-successional vegetation changes after roadside prairie restoration modify processes related with soil functioning by changing microbial functional diversity. Soil Biol Biochem 43:1245–53.

García-Palacios P, Milla R, Delgado-Baquerizo M, Martín-Robles N, Álvaro-Sánchez M, Wall DH. 2013. Side-effects of plant domestication: ecosystem impacts of changes in litter quality. New Phytol 198:504–13.

Giorgi F. 2006. Climate change hot-spots. Geophys Res Lett 33:L08707.

Halverson LJ, Jones TM, Firestone MK. 2000. Release of intracellular solutes by four soil bacteria exposed to dilution stress. Soil Sci Soc Am J 64:1630–7.

Harper CW, Blair JM, Fay PA, Knapp AK, Carlisle JD. 2005. Increased rainfall variability and reduced rainfall amount decreases soil CO_2 flux in a grassland ecosystem. Glob Chang Biol 11:322–34.

Hobbie SE, Oleksyn J, Eissenstat DM, Reich PB. 2010. Fine root decomposition rates do not mirror those of leaf litter among temperate tree species. Oecologia 162:505–13.

Jackson RB, Mooney HA, Schulze ED. 1997. A global budget for fine root biomass, surface area, and nutrient contents. Proc Natl Acad Sci USA 94:7362–6.

Kipfer T, Wohlgemuth T, van der Heijden MGA, Ghazoul J, Egli S. 2012. Growth response of drought-stressed *Pinus sylvestris* seedlings to single- and multi-species inoculation with ectomycorrhizal fungi. PLoS ONE 7(4):e35275.

Limousin JM, Rambal S, Ourcival JM, Rocheteau A, Joffre R, Rodríguez-Cortina R. 2009. Long-term transpiration change with rainfall decline in a Mediterranean Quercus ilex forest. Glob Chang Biol 15:2163–75.

Limousin JM, Rambal S, Ourcival JM, Rodríguez-Calcerrada J, Pérez-Ramos IM, Rodríguez-Cortina R, Misson L, Joffre R. 2012. Morphological and phenological shoot plasticity in a Mediterranean evergreen oak facing long-term increased drought. Oecologia 169:565–77.

Marigo G. 1973. Sur une méthode de fractionnement et d'estimation des composés phénoliques chez les végétaux. Analusis 2:106–10.

McClaugherty CA, Aber JD, Melillo JM. 1982. The rate of fine roots in organic matter nitrogen budgets of two forest ecosystems. Ecology 63:1481–90.

Misson L, Rocheteau A, Rambal S, Ourcival JM, Limousin JM, Rodríguez-Cortina R. 2010. Functional changes in the controls of carbon fluxes after 3 years of increased drought in a Mediterranean evergreen forest? Glob Change Biol 16:2461–575.

Norby RJ, Ledford J, Reilly CD, Miller NE, O'Neill EG. 2004. Fine-root production dominates response of a deciduous forest to atmospheric CO_2 enrichment. Proc Natl Acad Sci USA 101:9689–93.

Ogaya R, Peñuelas J. 2006. Contrasting foliar responses to drought in Quercus ilex and Phillyrea latifolia. Biol Plant 50:373–82.

Pérez-Harguindeguy N, Díaz S, Garnier E, Lavorel S, Poorter H, Jaureguiberry P, Bret-Harte MS, Cornwell WK, Craine JM, Gurvich DE, Urcelay C, Veneklaas EJ, Reich PB, Poorter L, Wright IJ, Ray P, Enrico L, Pausas JG, De Vos AC, Buchmann N, Funes G, Quétier F, Hodgson JG, Thompson K, Morgan HD, Ter Steege H, Van Der Heijden MGA, Sack L, Blonder B, Poschlod P, Vaieretti MV, Conti G, Staver AC, Aquino S, Cornelissen JHC. 2013. New handbook for standardised measurement of plant functional traits worldwide. Aust J Bot 61:167–234.

Pesaro M, Widmer F, Nicollier G, Zeyer J. 2003. Effects of freeze–thaw stress during soil storage on microbial communities and methidathion degradation. Soil Biol Biochem 35:1049–61.

Porter LJ, Hrstich LN, Chan BG. 1986. The conversion of procyanidins and prodelphinidins to cyanidin and delphinidin. Phytochemistry 25:223–30.

R Development Core Team. 2011. R: a language and environment for statistical computing. Vienna: R Foundation for Statistical Computing.

Rambal S, Ourcival JM, Joffre R, Mouillot F, Nouvellon Y, Reichstein M, Rocheteau A. 2003. Drought controls over conductance and assimilation of a Mediterranean evergreen ecosytem: scaling from leaf to canopy. Glob Change Biol 9:1813–24.

Richard F, Roy M, Shahin O, Sthultz C, Duchemin M, Joffre R, Selosse MA. 2011. Ectomycorrhizal communities in a Mediterranean forest ecosystem dominated by *Quercus ilex*: seasonal dynamics and response to drought in the surface organic horizon. Ann Forest Sci 68:57–68.

Sala OE, Chapin FS, Armesto JJ, Berlow E, Bloomfield J, Dirzo R, Huber-Sanwald E, Huenneke LF, Jackson RB, Kinzig A, Leemans R, Lodge DM, Mooney HA, Oesterheld M, Poff NL, Sykes MT, Walker BH, Walker M, Wall DH. 2000. Global biodiversity scenarios for the year 2100. Science 287:1770–4.

Sanaullah M, Chabbi A, Charrier X, Rumpel C. 2012. How does drought stress influence the decomposition of plant litter with contrasting quality in a grassland ecosystem? Plant Soil 353:277–88.

Sardans J, Peñuelas J. 2005. Increasing drought decreases phosphorus availability in an evergreen Mediterranean forest. Plant Soil 267:367–77.

Sardans J, Peñuelas J. 2007. Drought changes phosphorus and potassium accumulation patterns in an evergreen Mediterranean forest. Funct Ecol 21:191–201.

Saura-Mas S, Estiarte M, Peñuelas J, Lloret F. 2012. Effects of climate change on leaf litter decomposition across post-fire plant regenerative groups. Environ Exp Bot 77:274–82.

Sheffield J, Wood EF. 2008. Projected changes in drought occurence under future global warming from multi-model, multi-scenario, IPCC AR4 simulations. Clim Dyn 31:79–105.

Somot S, Sevault F, Deque M, Crepon M. 2008. 21st century climate change scenario for the Mediterranean using a couple atmosphere-ocean regional climate model. Glob Planet Change 63:112–26.

Terradas J. 1999. Holm oaks and holm oak forests: an introduction. In: Roda F, Retana J, Gracia CA, Bellot J, Eds. Ecology of Mediterranean evergreen oak forests. Berlin: Springer. p 3–14.

Violle C, Enquist BJ, McGill BJ, Jiang L, Albert C, Hulshof C, Jung V, Messier J. 2012. The return of the variance: intraspecific variability in community ecology. Trends Ecol Evol 27:244–52.

Vogel A, Eisenhauer N, Weigelt A, Scherer-Lorenzen M. 2013. Plant diversity does not buffer drought effects on early-stage mass loss rate and microbial processes. Glob Change Biol 91:485–96.

Walter J, Hein R, Beierkuhnlein C, Hammer V, Jentsch A, Schädler M, Schuerings J, Kreyling J. 2013. Combined effects of multifactor climate change and land-use on decomposition in temperate grassland. Soil Biol Biochem 60:10–18.

Wang H, Liu S, Mo J. 2010. Correlation between leaf litter and fine root decomposition among subtropical tree species. Plant Soil 335:289–98.

Xiang SH, Doyle A, Holden PA, Schimel JP. 2008. Drying and rewetting effects on C and N mineralization and microbial activity in surface and subsurface California grassland soils. Soil Biol Biochem 40:2281–9.

Zhang D, Hui D, Luo Y, Zhou G. 2008. Rates of litter decomposition in terrestrial ecosystems: global patterns and controlling factors. J Plant Ecol 1:85–93.