ARTICLE IN PRESS

Pedobiologia - Journal of Soil Ecology xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Pedobiologia - Journal of Soil Ecology

journal homepage: www.elsevier.com/locate/pedobi

Weak to no effects of litter biomass and mixing on litter decomposition in a seasonally dry tropical forest

Anderson R. Gripp[a,*], Francisco de Assis Esteves[a,b], Luciana S. Carneiro[c], Rafael D. Guariento[d], Marcos Paulo Figueiredo-Barros[b], Sylvain Coq[e], Alexandru Milcu[e,f], Adriano Caliman[c]

[a] Departamento de Ecologia, Universidade Federal do Rio de Janeiro, Prédio do CCS – Bloco A – Cidade Universitária – Ilha do Fundão, Caixa Postal 68.020, Rio de Janeiro, RJ, CEP. 21.941-540, Brazil

[b] Núcleo de Pesquisas em Ecologia e Desenvolvimento Socio-Ambiental de Macaé (NUPEM), Universidade Federal do Rio de Janeiro (UFRJ), Av. São José do Barreto, No. 764. Bairro São José do Barreto, Macaé, RJ, CEP: 27965-045, Brazil

[c] Departmento de Ecologia, Universidade Federal do Rio Grande do Norte (UFRN), Campus Lagoa Nova S/N, Centro de Biociências, Natal, RN, CEP 59078-900, Brazil

[d] Laboratório de Ecologia, Instituto de Biociências (INBIO), Universidade Federal do Mato Grosso do Sul (UFMS), Cidade Universitária S/N, Campo Grande, M. Caixa Postal: 549*, CEP: 79070-900, Brazil

[e] CEFE UMR 5175, Centre National de la Recherche Scientifique (CNRS), Université de Montpellier, Universite Paul-Valéry Montpellier, EPHE, IRD, 1919 Route de Mende, F-34293 Montpellier Cedex 5, France

[f] Ecotron Européen de Montpellier, Unité Propre de Service 3248, Centre National de la Recherche Scientifique (CNRS), Campus Baillarguet, F-34980 Montferrier-sur-Lez, France

ARTICLE INFO

Keywords:
Non-additive effects
Ecosystem functioning
Nutrient recycling
Restinga
Inter-specific litter interactions

ABSTRACT

Leaf litter mixtures and the amount of litter biomass in the litter standing stocks can affect the decomposition rates by modifying physical properties and resource heterogeneity in the litter layers. However, the potential interactive effects of litter mixtures and the amount of litter biomass on decomposition have been overlooked in the literature, even though both aspects of litter layer may be highly variable in space and time, within and across ecosystems. In a field experiment conducted in a seasonally dry tropical forest (also known as *restinga* forest), we investigated the individual and interactive effects of litter mixing and litter biomass on the decomposition of leaf litter from four species of trees, at both species- and assemblage-level. We hypothesized that the mixing of litter and higher litter biomass would both promote litter decomposition, and that litter mixture effects on decomposition at assemblage- and species specific-level would be stronger under higher litter biomass. Mixing of litter and the amount of litter in the standing stocks had no significant individual effects on decomposition, neither at the assemblage- or species specific-level. However, we observed an interactive effect between both experimental factors for the decomposition of a single species, where, contrary to our predictions, the decomposition of *Andira legalis* at low litter biomass was slightly reduced in the litter mixture. Our results indicate that litter decomposition in the *restinga* ecosystem should be highly predictable with knowledge of species composition and species-specific decomposition rates, and suggest that, at least for seasonally dry tropical ecosystems, the mixing of litter may have only a small effect on the decomposition process.

The decomposition of litter is a fundamental biogeochemical process controlling the nutrient, carbon and energy cycling in ecosystems (Parton et al., 2007; Swift et al., 1979; Wardle, 2002). There is unequivocal evidence that alongside climate, the quality (chemical and physical composition) and quantity of litter controlling the activity of decomposer communities are the main drivers of litter decomposition at large spatial scales (Handa et al., 2014; Parton et al., 2007; Swift et al., 1979). However, understanding why litter containing leaves from several species (hereafter, litter mixtures) decompose in a non-additive way compared to monospecific litter remains a major challenge.

One potentially important driver which has been shown to positively affect litter decomposition (Fang et al., 2015; Liu et al., 2016; Sayer et al., 2007; but see Parsons et al. (2014) for negative effects of litter biomass on decomposition) but has been overlooked in the context of non-additive litter mixing effects is the amount of litter biomass in the litter standing stock (hereafter, litter biomass). The discrepancies in litter quality present in mixtures may play a more important role where more litter accumulates due to, for example, time-mediated

* Corresponding author.
E-mail addresses: argripp@ufrj.br (A.R. Gripp), festeves@biologia.ufrj.br (F.d.A. Esteves), lscarnei@gmail.com (L.S. Carneiro), rafaguariento@gmail.com (R.D. Guariento), mpaulo.bio@gmail.com (M.P. Figueiredo-Barros), Sylvain.Coq@cefe.cnrs.fr (S. Coq), alex.milcu@ecotron.cnrs.fr (A. Milcu), caliman@cb.ufrn.br (A. Caliman).

https://doi.org/10.1016/j.pedobi.2018.02.003
Received 1 August 2017; Received in revised form 3 February 2018; Accepted 21 February 2018

Please cite this article as: Gripp, A., Pedobiologia - Journal of Soil Ecology (2018), https://doi.org/10.1016/j.pedobi.2018.02.003

vertical differences in the decomposition stage across thicker litter layers. Chemical dissimilarity (Epps et al., 2007; Hättenschwiler, 2005; Hättenschwiler and Jørgensen, 2010) and its impact on the selective feeding of decomposers (Swan and Palmer, 2006a,b) has also been demonstrated to play a role in litter mixtures effects. These selective feeding may depend on the proportion of high quality detritus in mixtures, and therefore, strongly depend on the amount of litter biomass in the standing stock. Moreover, as litter species differ in their physical properties, litter mixtures tend to have different microclimatic characteristics (Swift et al., 1979; Wardle et al., 1997) and habitat structure (Hättenschwiler et al., 2005; Makkonen et al., 2013), thereby altering (and often enhancing) litter decay rates (Wardle et al., 2003). The impact of litter biomass on the chemical, physical, micro-environmental and biotic changes in the environment affecting litter decomposition suggests that it may play an important role in the non-additive effects of litter mixtures. However, the impact of litter biomass on the decomposition of litter mixtures has not been yet assessed and remains an important gap in our knowledge.

To address this knowledge gap, we set up a litter decomposition experiment in a seasonally dry tropical forest to test the occurrence of a potential interaction between litter biomass and the decomposition of litter in two scenarios: monospecific and a four-species mixture. We hypothesized that i) both higher litter biomass and litter mixing will enhance the decomposition; and ii) the effect of litter mixing will depend on the amount of litter biomass in the standing stocks. The second hypothesis is based on the conjecture that changes in litter quality due to mixing may play a more important role where more litter is available, leading to more pronounced non-additive litter mixing effects in the treatment with higher litter biomass.

The experiment was carried out in a pristine seasonally dry tropical forest located at the Restinga de Jurubatiba National Park in the northern region of Rio de Janeiro state, Brazil (22°–22°30′S and 41°15′–45°W). The *restinga* ecosystem is part of the Atlantic Forest biome and is characterized by a mosaic of forests and shrublands growing on oligotrophic sandy soils, poor in clay and organic material, with a low water and nutrients retention capacity (Hay and Lacerda, 1984). The climate is seasonal tropical sub-humid to humid (1,165 mm $year^{-1}$), with the average (from years 2000 to 2009) of minimum precipitation occurring in winter (ca. 40 mm) and maximum in summer (ca. 190 mm) and a mean annual temperature of 22.6 °C (Caliman et al., 2010). Within the *restinga* mosaic, we selected four areas of seasonally dry forest occupying a fringe of about 30 m around Jurubatiba coastal lagoon (Supplementary Fig. S1; see below for more details). The experimental sites were not flooded during the experiment.

Restinga vegetation is mainly composed of species producing low quality and highly recalcitrant leaf litter, with strong leaf functional similarity due to structural and physiological adaptations to drought (Rosado and De Mattos, 2010). We selected leaf litter from four species commonly found in the study site (Pimentel et al., 2007) and exhibiting a gradient of degradability, based on litter breakdown rates (expressed in k ± standard deviation; $year^{-1}$) estimated in a previous trial experiment from a pool of 25 dominant tree species: *Andira legalis* (Vell.) Toledo (0.282 ± 0.068 y^{-1}), *Eugenia umbelliflora* Berg (0.391 ± 0.015 y^{-1}), *Clusia hilariana* Schltdl. (0.648 ± 0.061 y^{-1}) and *Schinus terebinthifolius* Raddi (1.093 ± 0.177 y^{-1}). Senescent leaf litter with less than 50% of green-chlorophylled surface (Supplementary Fig. S2) was collected from the trees just before they were shed as litterfall and oven-dried at 40 ± 5 °C until they reach a constant weight. Using a factorial randomized block design (with four blocks), we established a litter richness treatment (two levels – monospecific litter belonging to the 4 species and a 4-species mixture) and a litter biomass treatment (two levels – low and high, corresponding approximately to 225 g and 900 g of leaf litter dry mass per square meter, respectively) as orthogonal factors. Using a plastic cylindric sampler, we previously estimated the amounts of litter in the standing stocks using 12 transects with 3 sampling plots perpendicular to Jurubatiba lagoon in each transect. We took all the organic material inside the samplers to the lab and separated gross- and fine-fragmented leaves, dried at 40 ± 5 °C and weighted. The amounts considered in this experiment correspond to one-fourth and mean total natural leaf accumulated in the litter standing stock, respectively. The experiment was replicated in four spatial blocks, each block representing a branch around Jurubatiba lagoon (Supplementary Fig. S1). Each single species and 4-species mixture were replicated one and two times per block, respectively, for a total of 48 experimental replicates ([1 replicate per each of four species monoculture + 2 replicates for species mixture] × 2 levels of litter biomass × 4 spatial blocks). In the 4-species mixture each species contributed 25% of the total leaf litter in each replicate. In each block, we estimated top soil (0–5-cm depth) moisture by the gravimetric method and determined soil pH in water, according to Carter and Gregorich (2007). We also measured litter bed height and temperature, with a ruler and a digital thermometer (Minipa, model MV-363), respectively, in three sub-replicates within each block and mean values were registered (Supplementary Table S1).

Approximately 4 g and 16 g (weighted to the nearest 0.001 g) of leaf litter dry mass (i.e. low and high litter biomass, respectively) were added to plastic cylindric mesocosms (15 cm diameter, 10 cm height, and 0.0177 m^2 surface area), distributed randomly within each block and separated from each other by at least 25 cm (Supplementary Fig. S3). The mesocosms were covered with a 0.5 mm mesh at the top and the bottom to allow the passage of water while preventing the entrance of litterfall from local vegetation and the loss of small litter particles through the bottom. Two windows of 5 cm height and 18 cm wide, from approximately 1 cm above the microcosms' bottom, were made on opposite sides of the microcosms' wall, and covered with 5 mm mesh, following Handa et al. (2014). Visual inspection as well as soil fauna collected from a parallel experiment using the same mesocosms (unpublished data) indicate that macro- and mesofauna were able to access the mesocosms.

The experiment lasted 277 days from 03 July 2015 to 04 April 2016. At the end of the experiment, the remaining litter was oven-dried at 40 ± 5 °C to constant weight, cleaned by gently brushing and weighted. Litter mixtures were separated into the constituent species in order to assess species-specific effects; unidentified litter accounted for less than 5% of total mass.

Decomposition, both at assemblage and species-specific level, were expressed as the percentage of litter mass remaining. Data were analyzed using a mixed- effects modeling approach considering the "*lme*" function from the "*nlme*" package as available in R v. 3.3.3 (Team, R.D.C., 2017), following the recommendations of Zuur et al. (2009). The effect of litter mixture, litter biomass and their interaction on the response variables (assemblage and species-specific litter mass remaining) were tested as fixed factors, with experimental block as a random factor and a 'varIdent' weighting function to correct for heteroscedasticity due to not homogenous variance levels among the different treatment combinations (R syntax: 'model = lme (response variable ~richness*biomass, random = ~1|block, weights = varIdent (form = ~1|richness)'); see Supplementary Fig. S4 for a residual inspection of the fitted models. Data were *logit* transformed previous to analysis (Warton and Hui, 2011).

No statistically significant effect of litter mixture, litter biomass or their interaction was found on leaf litter mass remaining at the assemblage level (Table 1; Fig. 1a). At species level, for three out of the four litter species (*E. umbelliflora*, *C. hilariana* and *S. terebinthifolius*) we did not observe any significant statistical effects of litter mixture, litter biomass or their interaction on leaf litter mass remaining (Fig. 1). However, a significant interaction was observed between these factors on leaf litter mass remaining for *A. legalis* ($F_{1,16} = 7.37$, $p = 0.015$; Table 1), exhibiting a significantly slower (ca. 7.7%) decomposition in litter mixture than in monoculture in the low litter biomass treatment (Fig. 1e).