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Breaking down the effect of biotic and abiotic mechanisms of litter decomposition in drylands

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Breaking down the effect of biotic and abiotic mechanisms of litter decomposition in drylands

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Abstract

Plant litter decomposition constitutes one of the largest fluxes in the global carbon cycle releasing ≈68Pg C y^{-1} into the atmosphere. In arid and semi-arid systems, which account for ≈43% of the world's land area, litter decomposition rates are systematically underestimated by up to 30%, leading to a large part of the carbon budget being unaccounted for. Recent research has highlighted some of the potential mechanisms which lead to this underestimation but fails to elucidate how the various mechanisms interact. I investigated this by utilizing a fully-factorial experimental spanning both the dry and wet seasons, manipulating UV by filtering, temperature and humidity using Open Top Chambers and standardising wet season water input by spraying with deionised water. I demonstrate that, in the absence of precipitation, abiotic degradation (chiefly photodegradation, thermal decomposition and leaching) throughout the dry season contribute significantly to litter decomposition with litter mass loss of 60%. Photodegradation forms both diurnal and seasonal feedback-loops with microbial activity which are either sustained by night-time humidity/dew adsorption, rainfall or (artificial) watering. I estimated that the main mechanisms of litter decomposition over the dry period are thermal degradation that contributed more than 50% to litter mass loss while photodegradation contributed only 10%. The combined thermal and fungal degradation led to substantial decomposition of the soluble cell fraction (the most labile carbon) (59.9 ± 0.6% reduction). Despite the small contribution of photodegradation to overall decomposition, exposure to UV light led to a significant reduction in hemicellulose content by 26.30%, but had only a small, non-significant effect on mass loss (3.69%). The results indicate, that despite reduced dry season microbial decomposition in the filtered treatments there was a seasonal priming effect due to UV light exposure. Besides priming effects, dry season decomposition caused a shift in the fractions of the cell being decomposed. Due to the ubiquitous consumption of labile carbon throughout the dry season and the shift in dominant processes from abiotic to biotic, wet season

decomposition preferentially degraded the more recalcitrant compounds such as hollocellulose. Warming led to an increase in microbial decomposition by 26%, in photodegradation by 3% and a decrease in the relative influence of thermal decomposition by 28%.

This study finds that the unexpectedly high dry season relative decomposition rate is a consequence of the strength of the feedback loops between abiotic and biotic mechanisms of decomposition. It highlights the necessity to approach dryland litter decomposition with a integrative view if we are to accurately predict litter decomposition rates and estimate carbon budgets.

Keywords: Leaf litter decomposition, photodegradation, thermal degradation, semi-arid ecosystems, microbial degradation, microbial priming, warming manipulation, litter structural changes, enzymatic activities

INTRODUCTION

Plant litter decomposition links aboveground and belowground processes and is essential for carbon and nutrient turnover in terrestrial ecosystems (Cotrufo *et al.*, 2013). Together with soil organic matter decomposition (heterotrophic respiration), litter decomposition constitutes one of the largest fluxes of the global carbon cycle (Andrews & Schlesinger, 2000). As a result of soil microbial decomposition of litter and soil organic matter ≈68Pg C y^{-1} are released to the atmosphere (Raich & Schlesinger, 1992; Baker & Allison, 2015) compared to ≈6Pg via fossil fuel burning (Denman & Hauglustaine, 2007; Bonan *et al.*, 2013). In terrestrial ecosystems, more than 50% of net primary production is returned to soils via the decomposition of leaf litter (Wardle *et al.*, 2004). Therefore, understanding the factors controlling litter decomposition is crucial for the quantification of present and future global carbon budgets (Aerts, 2006).

The main factors controlling litter decomposition are climate, litter quality, and soil biota (Berg & McClaugherty, 2008). Biotic litter decomposition involves a range of soil organisms, including invertebrates and microbes, which oxidise large quantities of plant organic matter as a source of energy and nutrients. Climate exerts a direct control on litter decay since biological activity strongly depends on temperature and precipitation (Berg & Staaf, 1980). In addition, climate also indirectly affects litter decomposition through its effects on litter quality and soil decomposing biota (Wardle *et al.*, 2004). Decomposer activity is controlled by environmental factors (i.e., temperature and water availability) and litter quality (Gavazov, 2010), including lignin and nitrogen content (Couteaux *et al*., 1995; Cornwell *et al.*, 2008; Incerti *et al.*, 2011; García-Palacios *et al.*, 2013). Litter quality is an important factor given that labile components are more easily degradable than recalcitrant ones. The effects of climate and litter quality upon litter decomposition rates have been previously evaluated at both regional and global scales (e.g. Swift *et al*. 1979; Couteaux *et al*. 1995; Aerts 1997; Cornwell *et al*. 2008; García-Palacios *et al.*, 2013). However, recent biogeochemical models using climate and litter quality explain

about 60–70% of global litter decomposition rates and systematically underestimate litter decomposition in drylands (Parton *et al.*, 2007; Adair *et al.*, 2008; Bonan *et al.*, 2013). Given the large proportion of global land area (43%) and carbon stocks (≈21%) of drylands (Safriel *et al.*, 2005), these ecosystems play a crucial role in global carbon cycling and control interannual variability in the global carbon budget (Ahlström *et al*., 2015). It is therefore essential to improve the understanding of litter decomposition processes in drylands.

Traditional exponential decomposition models state that precipitation is a core driver of litter decomposition (Vernon Meentemeyer, 1978) and that decomposition is predominantly microbially mediated (Paudel *et al.*, 2015). However, in arid ecosystems, microbes can be activated by relative humidity (RH) (Dirks *et al.*, 2015; Gliksman *et al.*, 2018) and thus microbial activity may be underestimated when precipitation is assumed to be the only source of water. The fact that current models consistently fail to predict litter decay rates in arid and semiarid ecosystems (Whitford, 1981; Parton *et al.*, 2007) suggests that other mechanisms and factors contribute to litter decomposition in these water-limited ecosystems (Throop & Archer, 2009; King et al., 2012). Recent research has shown that abiotic processes of litter decomposition contribute considerably to the degradation of leaf litter in such ecosystems (e.g. King *et al*., 2012; Barnes *et al.*, 2015; Gliksman *et al.*, 2017; Lin *et al.*, 2018). Abiotic processes include photodegradation (the degradation of litter by solar exposure) (Austin & Vivanco, 2006; Brandt, Bonnet and King, 2009; Lee, Rahn & Throop, 2012; Whelan & Rhew, 2015; Lin *et al.*, 2018) and thermal degradation (litter degradation at temperatures above 30˚C) (Lee *et al*., 2012; Whelan & Rhew, 2015; Gliksman *et al.*, 2017). Moreover, relative humidity has also been demonstrated to be an important factor controlling biological activity in drylands (Dirks *et al*, 2015; Gliksman *et al*., 2018). Photodegradation is the process by which solar radiation directly breaks down organic matter components releasing CO_2 (Van Asperen *et al.*, 2015). Thus, it constitutes a direct loss of carbon from ecosystems to the atmosphere without being

incorporated into the soil organic matter pool (Austin & Vivanco, 2006). A growing body of literature has shown that solar ultraviolet (UV) radiation (280-400 mm) can be an important driver of leaf litter decomposition in semiarid ecosystems (e.g. Brandt *et al*. 2007; Day *et al*. 2007; Gallo *et al*. 2009; Rutledge *et al*. 2010; Baker & Allison 2012; Gliksman *et al*. 2017; Huang *et al*. 2017; Lin *et al*. 2018). A meta-analysis concluded that solar radiation speeds up decomposition by 32% (King *et al*., 2012) but results differ among litter types, site characteristics (solar irradiance, temperature, moisture, etc.) and experimental conditions (field vs. laboratory) (Gallo *et al*., 2006; Brandt *et al.*, 2010; Almagro *et al.*, 2015),. However, the magnitude and proposed mechanisms for this mass loss remain unclear (Song *et al.*, 2013; Barnes *et al.*, 2015).

Abiotic and biotic mechanisms seem to interact through a positive feedback (Gliksman *et al*., 2017) whereby photodegradation facilitates microbial decomposition by breaking down large organic compounds into smaller, more easily degradable (Austin & Ballare, 2010; Foereid *et al.*, 2010; Austin *et al*., 2016). Light, especially in the UV range, is absorbed by photochemically reactive regions in phenolic compounds such as lignin (Moorhead & Callaghan, 1994; Pancotto *et al.*, 2005; Austin & Ballare, 2010) leading to the breakdown of these macromolecules into smaller units that degrade or leach more easily or become more susceptible to microbial decomposition (photo-facilitation) (Austin *et al.*, 2016). In plants, lignin functions as a structural, hydrophobic barrier, shielding varying amounts of cellulose from microbial attack (Chen & Dixon, 2007; Gressel, 2008; Austin *et al.*, 2016). Lignin degrades when exposed to UV radiation, especially blue-green light, increasing subsequent microbial decomposition due to increased access to plant-litter carbohydrates (photo-priming) (Austin *et al*., 2016; Lin *et al.*, 2018). The combination of photo-priming and facilitation form a biotic-abiotic feedback loop (Figure 1) by which the increased microbial decomposition boosts the impact of subsequent UV exposure (Austin *et al.*, 2016). Photodegradation enhances

litter solubility, increases leaching of dissolved organic carbon (Gallo *et al*., 2006) and promotes direct photochemical mineralisation of litter releasing CO_2 (Brandt *et al*., 2007; Lee *et al*., 2012). Austin and Vivanco (2006) found evidence for an alternate mechanism of photomineralisation whereby mass loss preferentially occurred from the labile fraction indicating that the process is dynamic and could be mediated by environmental characteristics as well as by litter quality. Alternatively, solar radiation may influence microbial communities directly, either by inhibiting microbial activity and slowing down litter decomposition rates (Verhoef *et al.*, 2000; Pancotto *et al.*, 2003; Smith *et al.*, 2010) or by promoting changes in microbial diversity (Anesio *et al.*, 1999; Crutzen et al., 1999; Smith *et al.*, 2010). Microbial communities can be adapted to high UV environments (Brandt *et al.*, 2010) through the production of protective pigments (Gallo *et al.*, 2009). Thus, photodegradation is a complex process in which several mechanisms may interact. The effects of the interactions between UV radiation and other environmental factors such as humidity and temperature, are largely unknown and may vary depending on site-specific and climatic conditions, ecosystem type and land use (Gaxiola & Armesto, 2015; Almagro *et al.*, 2016; Huang & Li, 2017). Almagro *et al*. (2016) showed that the contribution of photodegradation to litter decomposition depends on environmental conditions with contrasting responses in continental and maritime climates and, suggest that photodegradation might interact with microbial decomposition (Figure 1). While most studies have focused on mass loss or carbon compounds, others have shown that photodegradation of specific plant components can lead to an increase in the release of N from litter, which in turn affects plant uptake, microbial communities, gaseous N emissions, and N leaching (McCalley & Sparks, 2009; Mayer *et al.*, 2012). To what extent carbon and nitrogen cycling are affected by these processes and will be affected by future climate change predictions remains unclear.

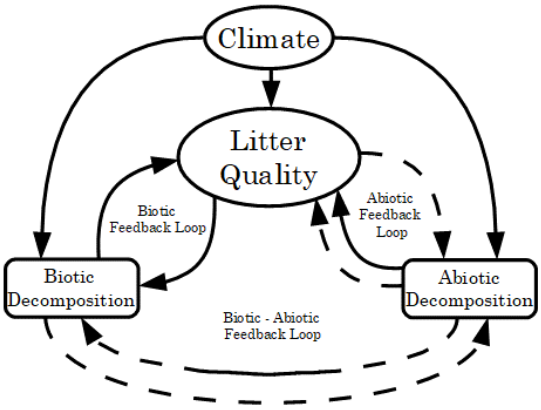

Figure 1. Conceptual model of the feedback loops underlying litter decomposition in drylands. Rectangles in the main pathways of decomposition and ellipses indicate the controlling factors of decomposition. Full arrows indicate direct effects, dashed arrows indicate indirect effects whilst full-dashed arrows indicate both direct and indirect effects being present.

Besides solar radiation, temperatures over 30˚C can also lead to litter mass losses through the preferential cleavage of chemical bonds of more recalcitrant organic materials (Lee *et al*., 2012; Almagro *et al.*, 2016) or via their reaction with reactive compounds through a process known as thermal degradation (Lee *et al*., 2012). Gliksman *et al*. (2017) estimated that thermal degradation contributes 12% to litter decomposition of Mediterranean grass species. However, changes in litter quality as a consequence of thermal degradation, the compounds released and how it interacts with microbial degradation are not known. The high temperatures associated with thermal decomposition have mixed effects on microbial communities. There is an exponential increase in the rate of enzyme reactions with increasing temperature up to a clearly definable optimum associated with the enzymes unusual heat capacity changes (Schipper *et al.*, 2014). Microbes in arid systems are adapted to the decreased soil moisture content associated with increasing temperature (Gliksman *et al.*, 2017) through the secretion of extracellular polymeric substances and gelatinous materials (Budel *et al.*, 2004; Warren-Rhodes *et al.*, 2007; Wong *et al.*, 2010; Pointing & Belnap, 2016).

Mediterranean ecosystems are particularly vulnerable to climate change (Sala *et al.*, 2000; Schröter *et al.*, 2005) as a result of reduced rainfall and changes in rainfall patterns (Lin *et al*.

2015). Current global circulation models predict an increase in temperature and reductions in precipitation for these regions (Giorgi & Lionello, 2008; IPCC, 2013). Due to the complex interactions between dryland litter decomposition and environmental factors such as temperature and humidity it is uncertain how climate change will impact litter decomposition in drylands. Therefore, the contribution of biotic and abiotic decomposition drivers to litter decomposition, and how they interact in semi-arid ecosystems remains unresolved, particularly during the long dry periods that characterise these biomes. Gliksman *et al*. (2017) studied litter composition in several grassland species in a Mediterranean transect over the dry season and identified a complex interaction between biotic degradation at night and abiotic degradation during daytime demonstrating that there is a positive feedback between both mechanisms. In this dissertation, I carried out an experiment to study the possible feedbacks mechanisms between abiotic and biotic mechanisms and the factors that control these processes.

The **objective** of this study was to investigate the processes involved in litter decomposition during the dry and wet seasons and the possible feedback mechanisms between abiotic and biotic processes of litter decomposition in drylands. In particular, I aimed to understand what structural, biochemical and microbial changes are caused by UV exposure over the dry season (summer), and how ultraviolet light exposure affects subsequent decomposition during the wet season (winter). Specific objectives were:

(1) To investigate the seasonal dynamics of dryland litter decomposition over a dry and wet season

(2) To investigate how exposure to solar radiation over the dry period affects subsequent litter decomposition during the wet season.

(3) To determine possible feedback mechanisms between abiotic and biotic processes of litter decomposition

(4) To determine changes in litter chemistry as a result of abiotic and biotic processes of litter decomposition

(5) To determine how increased temperature will impact litter decomposition by studying the impact of warming on litter decomposition in drylands.

It was hypothesized that (1) litter decomposition takes place during the dry season due to both the documented abiotic pathways such as photo and thermal degradation (*see* King *et al.*, 2012) and, as postulated by Gliksman *et al.* (2017), by night time microbial activity being driven by atmospheric moisture in periods with no rain; (2) the pathways of litter decomposition are expected to shift from abiotically mediated processes in the dry season to biotically mediated ones in the wet season; (3) decomposition over the wet season will be significantly higher for litter previously exposed to UV light than for shaded litter as a result of a positive feedback between abiotic and biotic degradation; (4) I expect will be clear evidence to support seasonal (Gallo *et al.*, 2009; Austin & Ballare, 2010) and diurnal photopriming (Gliksman *et al.*, 2017) and (5) that warming will reduce litter decomposition as a result of microbial inhibition through its indirect effect reducing relative humidity.

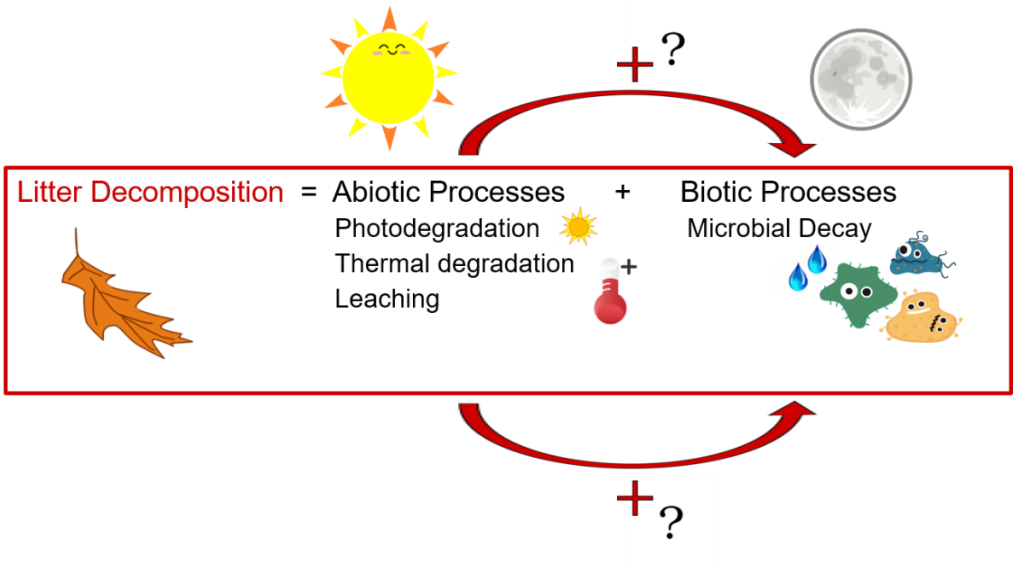

Figure 2. Aim of the study: structural and microbial changes as a result of biotic and abiotic processes and how abiotic processes modify subsequent biotic processes.

MATERIAL AND METHODS

Study site

A litter decomposition experiment was carried out at the Climate Change Outdoor Laboratory (CCOL) of King Juan Carlos University (URJC) located in Mostoles the community of Madrid, in the centre of the Iberian Peninsula (40°02´N, 3°32W; 590 m a.s.l). The Köppen-Geiger climate classification is Csa (Kottek *et al.*, 2006) and defined as semiarid Mediterranean, with mean annual temperature of 14.4 ˚C and rainfall of 449 mm (Almagro *et al.*, 2015). Rainfall follows a bimodal distribution with two rainy seasons (autumn and spring) and a dry period in summer with practically no rain (Almagro *et al.*, 2015).

Experimental design

With collaborators, I set up a three-way factorial experiment at the CCOL with the following treatments: (1) a *climatic manipulation treatment* with two levels: **CONTROL** (C) (ambient temperature) and **WARMING** (W) (a 3 ˚C annual temperature increase), (2) a *radiation manipulation treatment* with three levels: **control** (without radiation screens) [c], a **ultraviolet radiation transparent treatment** using filters that allow full radiation to pass [+UV], and a **radiation block treatment** (solar UV and shortwave PAR radiation blocked) [-UV] and (3) SEASON, a third factor (within subject) with two levels: **DRY** season (summer) and **WET** season (winter). Six replicates of each climatic treatment were randomly distributed in the study area and within each, the three radiation manipulation treatments were set up (Figure 3). Thus, the experimental design was a three-way split plot design with two between subject factors (climate and radiation) and one within subject factor (SEASON).

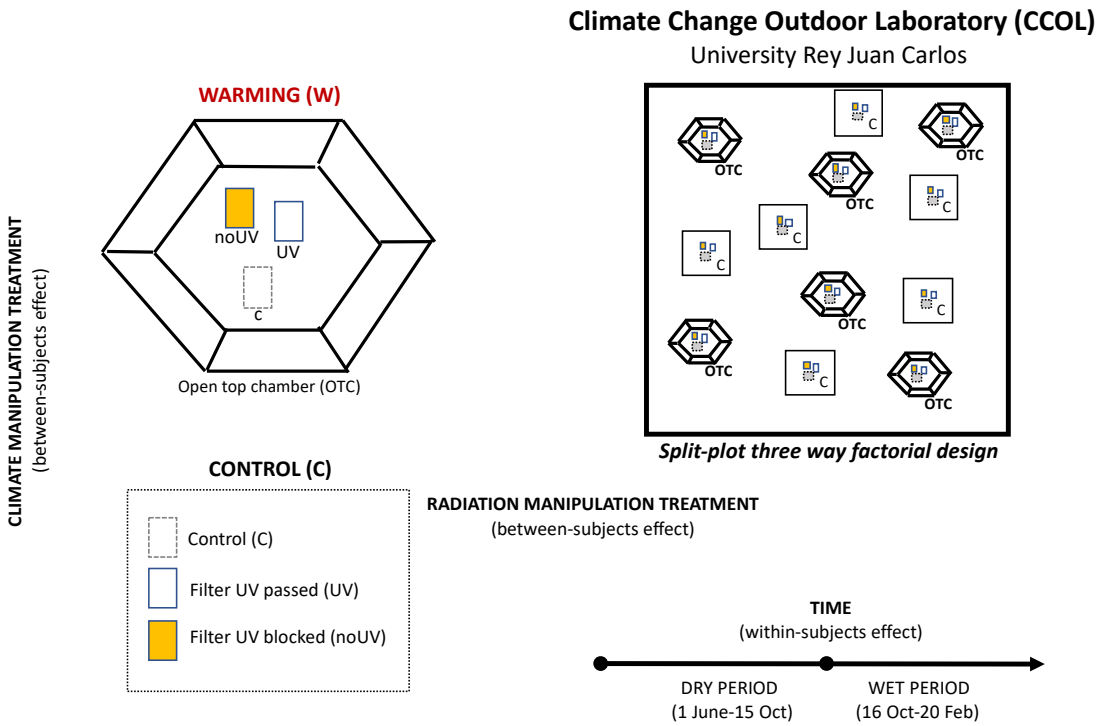

Figure 3. Experimental design at the Climate Change Outdoor Laboratory at the University Rey Juan Carlos.

Climate manipulation treatment

To achieve an annual increase in air temperature of 2-4 ºC, open top chambers (OTCs) were utilized as described by Maestre *et al*. (2013). The OTCs were built with six methacrylate plates, using a hexagonal design with sloping sides of 65 cm x 52 cm x 42 cm (Photo 1-2). This material was selected because of its very high transmittance of both visible (92%) and ultraviolet wavelengths (≥85%), and low reflection of incoming radiation (4%), allowing most of the incoming energy to pass (85%), while having a very low transmittance of infrared wavelengths (Maestre *et al.*, 2013). The OTCs were open at the top to allow precipitation and air to enter and were suspended 5 cm from the ground to allow air circulation and prevent overwarming (Almagro *et al.*, 2015) (Photo 1).

Photo 1. Open top chamber (OTC) used to obtain a 2-4ºC increase in temperature at the CCOL site.

Radiation manipulation treatment

The two radiation treatments were achieved by mounting filters as previously described. The UV pass filters (4000TR, Honeywell International, Morristown, NJ, USA) that allow near full radiation (92%) and filters (179 Chrome orange filters, Lee Filters, Burbank, CA, USA) that block all radiation below 550 nm. Each 30 cm x 30 cm filter screen was mounted with four screws and suspended 30 cm above the ground. All filters were placed south facing at the CCOL facility to ensure that they received the same amount of radiation (Photo 2).

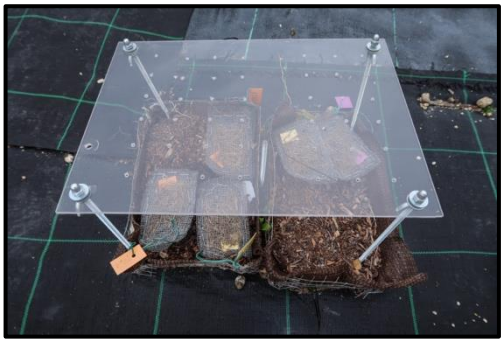

Photo 2. Deployment of radiation treatments within the OTCs.

Plant material and litterbag deployment

Recently senesced plant material was harvested from annual species and mixed: 80% *Festuca arundinacea*, 15% *Lolium perenne* and 5% *Poa pratensis.* The litter mix was selected due to its high degradability, characterised by a low C:N ratio and low lignin:cellulose index (LCI) which would increase the decomposition potential throughout the short time-span of the experiment allowing for the assessment of litter decomposition processes over the dry and wet seasons. This is a common mix of herbaceous species widely distributed across semiarid lands. Plant material was air dried until a constant mass was achieved and passed through a 4 mm sieve to remove small pieces. Dry litter was placed in 10 cm x 5 cm mesh bags (grey fiberglass at the bottom of 1.4 mm mesh size and transparent polyethylene with 90% transmittance of solar radiation at the top, 2 mm x 3 mm mesh size (Crystal, Meteor, Petah Tikva, Israel)). Each litterbag contained 0.765 (±0.055) g of litter. Eight litterbags were placed on replicated commercial garden soil pots made of a metal mesh of 20 x 12 and 5 cm deep (Photo 2), such that each sample consisted of approximately 6.28 g of grass litter material. The initial litter C and N content, lignin, hemicellulose and cellulose concentrations were determined at the beginning of the experiment using the methods described below (Table 1).

Table 1. Initial litter chemistry of the litter mix (80% *Festuca arundinacea*, 15% *Lolium perenne* and 5% *Poa pratensis*). Values are the mean ±1SE (n = 9 except for C, N and C:N where n = 6).

Component	**Mean** ±1SE
% Cell Soluble	51.54 ± 0.38
% Hemicellulose	20.50 ± 0.25
% Cellulose	25.44 ± 0.18
% Lignin	2.52 ± 0.07
% Carbon	41.90 ± 1.06
% Nitrogen	3.45 ± 0.10
% Ash	1.38 ± 0.08
% Water	6.00 ± 0.11
Lignin:cellulose (LCI)	0.09 ± 0.00
C:N ratio	13.05 ± 0.12

The experiment started on 1 June, 2017. The first set of litterbags was collected after 136 days, on the 14th of October, 2017, just before the first autumn rains started (after the DRY period) and the remaining bags were collected after 265 days, on 20 February, 2018, at the end of the experiment (after the WET period). The two samplings were designed to assess the effect of UV exposure during the dry season on subsequent litter decomposition over the wet season. To ensure equal amount of rain during the wet season in all treatments, all litterbags were watered weekly using deionised water with an amount of water equivalent to 5 mm of rain. Water was evenly applied with a sprayer over the litterbags. Eight litterbags were deployed per sample to ensure that there was enough material for the analyses and enough surface area would be exposed to light and air relative humidity to mimic natural conditions.

Monitoring of environmental variables

Air temperature and relative humidity were monitored 20cm above-ground using e-buttons Pro v2 U23-001 dataloggers (Onset, Bourne, MA, USA). Two sensors were used per combination treatment and air temperature and relative humidity were recorded at 30-minute intervals. UV radiation and PAR were also recorded using Ultraviolet meters (MU-200 and MQ-200, Apogee Instruments, Logan, Utah, USA). The effect of the UV filters and OTCs on UV radiation were measured on several occasions over the duration of the experiment (data not shown).

Litter mass loss

Three litterbags were collected after the DRY season on 15 October and five litterbags at the end of the experiment after the WET season on 15 February. All bags were weighed immediately after collection and litter mass loss on an ash-free basis was determined. A detailed methodology consisting of all protocols which had to be modified to account for the limited mass of litter are provided as supplementary material (S1). To determine litter moisture

content at the time of sampling, the fresh mass of all samples was determined. Litter samples were oven dried at 68°C for 48 h, after which dry mass was determined.

The litter remaining mass (RM) was calculated according to Wang *et al*. (2009):

$$\text{RM (g Ash-Free Dry Mass (AFDM))} = \frac{X_i}{X_0} \times 100$$

where:

X_0 = Initial ash free, dry litter mass

X_i = Ash free initial fresh mass when collected

Litter chemistry

The soluble cell fraction, hemicellulose, cellulose and acid-insoluble (lignin-like) (Corbeels, 2001) compound concentrations were determined by subjecting leaf litter to crude fibre analysis, using an Ankom Fiber Analyser (Ankom^{200} Fiber Analyzer, Ankom Technology Corp., Fairport, NY, and Fibertec I, Perstorp Analytical, Silver Spring, MD) followed by sulfuric acid digestion according to the standard protocol (Ankom Technology 2016; 2017a,b). Ground material (0.5 ± 0.05g) was subject to a series of sequential extractions in the order of: Neutral Detergent Fibre (NDF) (Ankom Technology, 2017b), Acid Detergent Fibre (ADF) (Ankom Technology, 2017a) and Acid Determined Lignin (ADL) (Ankom Technology, 2016). Ash content was subsequently determined by combustion in a muffle furnace (500 °C for 4h), ash included inorganic content and soil particles. Subsamples of litter (n = 4, 350 ± 100 mg) were ground to a fine powder by ball milling and analysed for total C and N using an elemental combustion analyser. From this, litter C to N, N to L and cellulose to lignin ratios were calculated.

Calculations of quantities of chemical constituents were carried out according to standard protocols (Ankom Technology 2016; Ankom Technology 2017 a; Ankom Technology 2017b; Wang *et al*. 2009*;* see supplement S1).

Extracellular enzyme activities

Litter samples were assayed for the potential activities of: β-D-xylosidase (X), β-1,4-N-acetylglucosaminidase (NAG), β-D-glucosidase (G), Cellobiohydrolase (CBH), acid phosphatase (AP), phenol oxidase (POX) and peroxidase (PER). All assays except POX and PER were fluorometric, utilizing 4-methyumbellifery (MUF) linked substrates (S2), the former being colorimetric, utilising L-3,4-dihydroxyphenylalanine (DOPA) as substrates. Assay preparation followed standard protocols (Saiya-cork *et al.*, 2002; Gallo *et al.*, 2006; Jackson *et al.*, 2013), which were tailored to this experiment (see supplement S1). Samples were frozen and transported at -80 °C and were processed within 24 h of defrosting. Samples were ground for 90 seconds in a ball mill grinder until they were a course powder. Subsequently, 0.5 g of litter was added to 100 ml of 5 mM, pH 5.0, acetate buffer (Saiya-cork *et al.*, 2002) and homogenised by constant stirring on a magnetic stir plate. Fluorometric assays were performed on black, 96-well microplates (Corning® 96 well NBS™ Microplate). Each microplate contained: 9 samples (well 1-9), three assay replicates (wells A-C) containing 200 µl of sample and 50 µl of model enzyme substrate, two sample control replicates (wells D-E) containing 200 µl of sample and 50 µl of buffer, three quench control replicates (wells F-H) containing 200 µl of sample and 50 µl of standard, three substrate control replicates (wells A10-A12) containing 200 µl of buffer and 50 µl of model enzyme substrate and three standard replicates (well H10-H12) containing 200 µl of buffer and 50 µl of standard. Aliquots of samples (200 µl) were dispensed onto the microplate whilst sample suspension was constantly stirred using 200 µl pipette tips (Eppendorf™ epT.I.P.S.™) with the heads cut off to prevent blockage and

minimise pipetting error. Enzyme activity (nmol h^{-1} g $AFDM^{-1}$) was calculated following German *et al*. (2011). All assays were optimized prior to analysis by utilizing "dummy-runs" where assays were performed using the standardized litter mix at varied substrate concentrations and incubation times (see S1).

Statistical analysis

A three-way ANOVA with three fixed factors: CLIMATE and UV treatments as between subject factors and SEASON as within subjects factors, was conducted using R statistical program (R Development Core Team, 2013). Given the large number of variables tested, *P* values were corrected with the false discovery rate correction to reduce the risk of Type-I error in the post-hoc t-tests. A variety of R packages were used throughout the analytical process (Venables & Ripley, 2002; Fox & Weiberg, 2011; R Core Team, 2013; Halekoh & Højsgaard, 2014; Bates *et al.*, 2015; Rosario-Martinez, 2015; Tremblay & Ransijn, 2015; Kuznetsova *et al*., 2017; Barton, 2018; Fox *et al*., 2018; Mangiafico, 2018; Revelle, 2018). To select the most appropriate models for analysis, Akaike information criteria (AICs) and Kenwoods-Rodgers statistics were calculated for null, full and no interaction models. The model with the lowest AIC was selected. Any data that showed a non-normal distribution or heteroskedasticity were either log(x+1), sqrt(x+1) or cubert(x+1) transformed, depending on whether they met parametric assumptions. Most instances of heteroskedasticity were rectified and most data were normalised, the exceptions being soluble cell fraction, phosphatase and xyloside which are approaching normality. Only C/N ratio showed a non-normal distribution and heteroskedasticity so a null-model was ran.

RESULTS

Effect of manipulation treatments on environmental conditions

During the DRY season, average temperature and humidity were 27.2 ± 0.05°C and 36.8 ± 0.1%, and during the WET season 8.8 ± 0.04°C and 74.9 ± 0.13%, respectively. Total precipitation throughout the DRY season was 52 mm and 171 mm throughout the WET season. Due to the predicted lack of rainfall throughout the WET season, all treatments were artificially watered, receiving the equivalent to 5 mm weekly so the total amount of rainfall received was 171 mm (Figure 4).

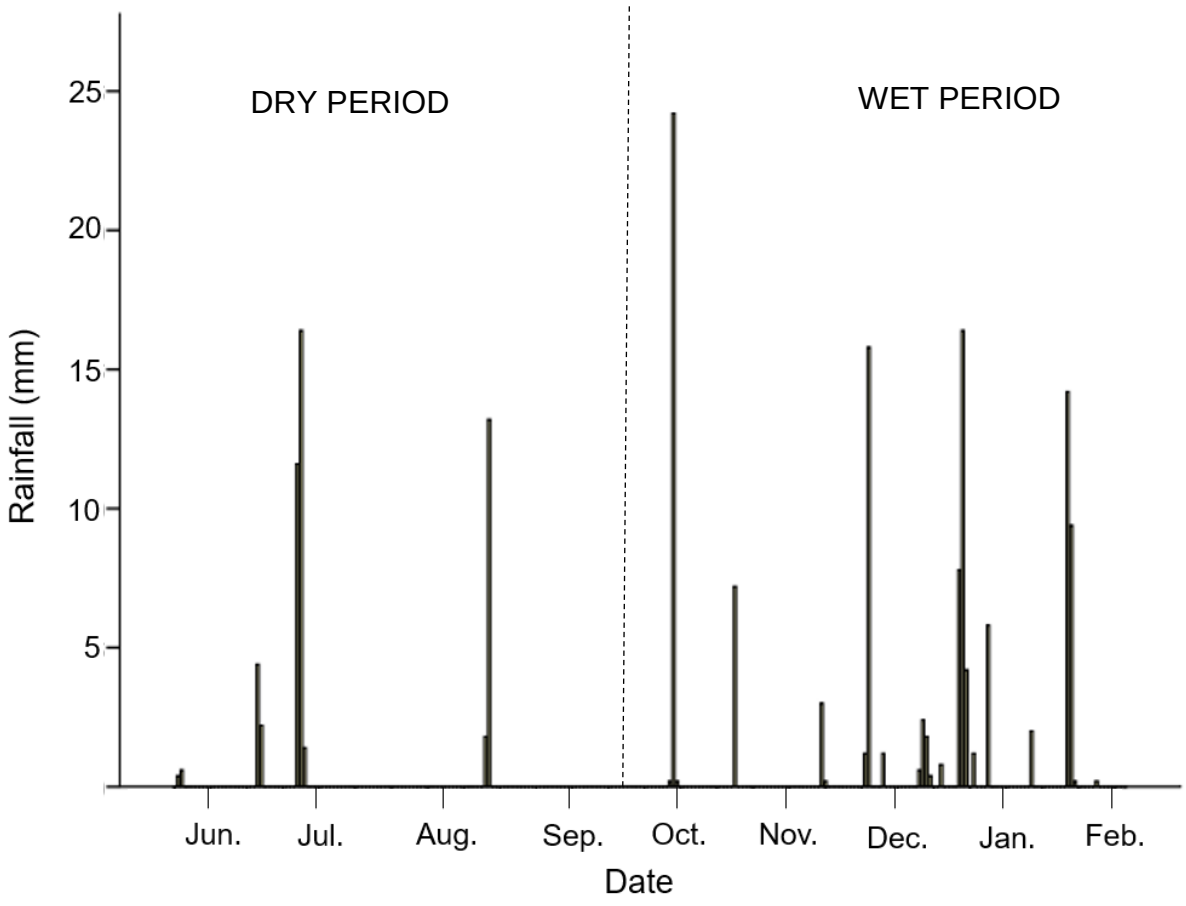

Figure 4. Natural precipitation events throughout the experimental period: DRY season (1/06/2017-19/10/2017) and WET season (20/10/2017-17/02/2018).

As the filters used for the different UV treatments were likely to intercept rainfall and reduce the free movement of air around the samples, it was expected that both air temperature and air relative humidity would be affected. Differences in mean air temperature and relative humidity were tested between seasons and among treatments. Introducing filters in the CONTROL treatment caused on average an increase in temperature of 0.79 ± 0.23°C and a decrease in air humidity of 1.71 ± 0.86% compared to the control treatment. The effect of filters varied depending on season, climate manipulation and day/night (Figure 5, Table 2).

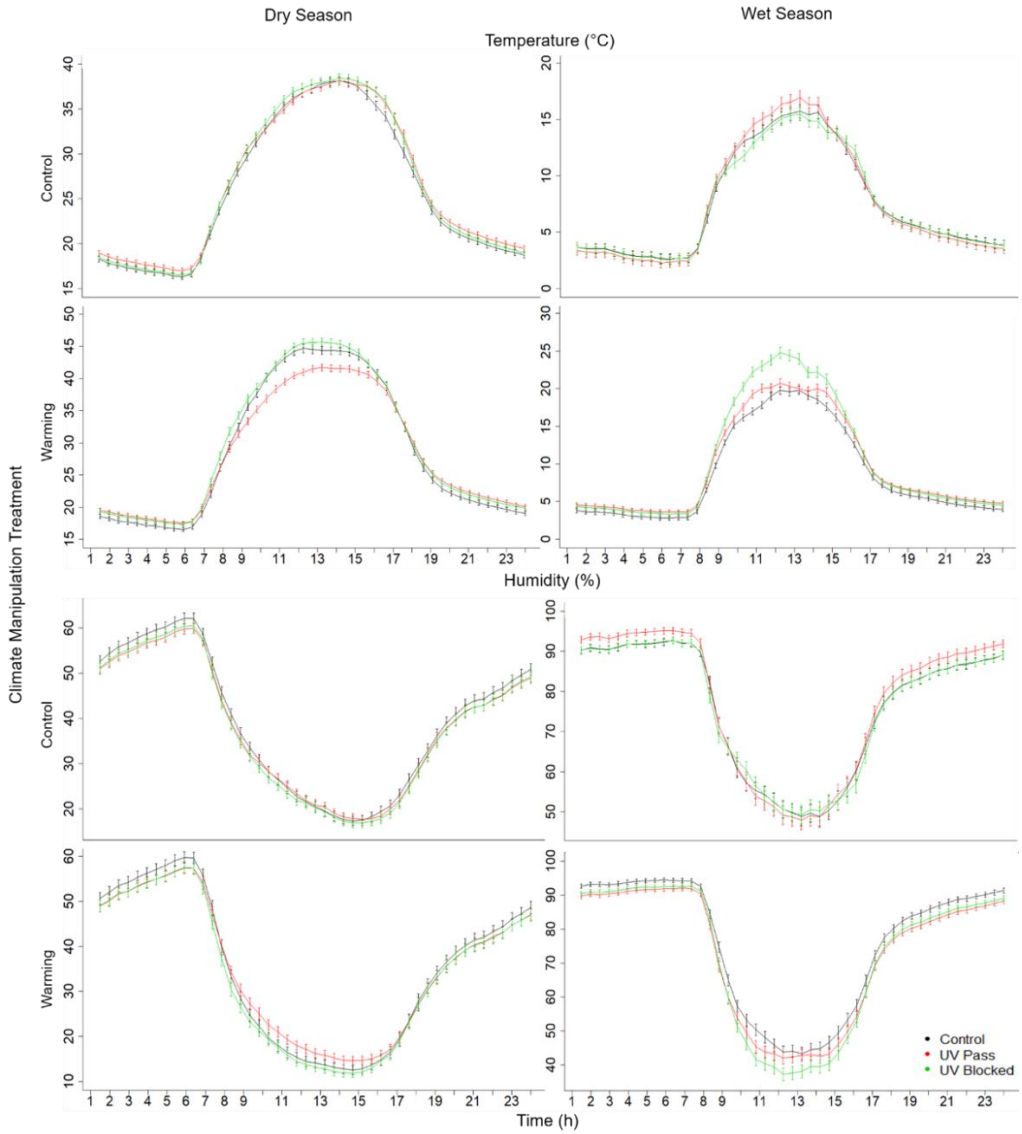

Figure 5. Diurnal cycles of air temperature (°C) and relative humidity (%), averaged throughout seasons, in the climate manipulation treatments (CONTROL and WARMING). The black, grey and dotted lines represent control, +UV and -UV treatments, respectively. Error bars represent ±1SE (*n* = 2).

The introduction of OTCs caused an overall increase in temperature of 2.16 ± 0.25°C and decrease in humidity of 3.26 ± 0.01% compared to the CONTROL treatment (Table 2). The effect of the open top chambers varied with season, UV manipulation and time of the day (Figure 5).

Table 2. Mean air temperature (°C) and relative humidity (%) in the climate manipulation treatments (CONTROL and WARMING) and radiation treatments (control +UV and -UV) in the DRY and WET seasons. Values are the mean ± 1SE (n = 2).

SEASON	Climatic treatment	Radiation treatment	Temperature (°C)	RH (%)
DRY	CONTROL	- UV	26.3 ± 0.1	38.2 ± 0.2
		+UV	26.2 ± 0.1	38.0 ± 0.2
		Control	25.7 ± 0.1	39.3 ± 0.2
	WARMING	- UV	27.8 ± 0.2	35.7 ± 0.2
		+UV	29.2 ± 0.1	33.9 ± 0.3
		Control	28.7 ± 0.1	34.8 ± 0.3
WET	CONTROL	- UV	7.5 ± 0.1	77.9 ± 0.4
		+UV	7.3 ± 0.1	76.2 ± 0.3
		Control	7.4 ± 0.0	76.3 ± 0.3
	WARMING	- UV	9.6 ± 0.1	73.1 ± 0.3
		+UV	10.2 ± 0.1	72.6 ± 0.3
		Control	8.6 ± 0.1	76.2 ± 0.3

Litter decomposition rate

Litter relative decomposition rates (k) varied between seasons, climate and radiation manipulation treatments (Table 3). Overall k was marginally, but significantly lower in the WET season ($F_{30,1} = 6.021$, $P < 0.05$), except for the UV treatments in the CONTROL treatment that were slightly faster in the WET season than in the DRY season. The UV manipulation treatment had the greatest effect on k ($F_{2,30} = 170.53$, $P < 0.001$), explaining 26% of the variation ($\eta^2 = 0.26$). Relative decomposition rate (k) was significantly higher (almost 100%) in all control plots compared to -UV and +UV treatments ($P < 0.001$ for all instances),

particularly in the DRY season. While litter decomposition in the +UV treatment was marginally higher (ca. 12%) than in the -UV treatment in all cases, differences between -UV and +UV treatments were not significant for any instances (Figure 6). Differences between radiation treatments were much larger in the DRY period than in the WET period (Figure 6).

Table 3. Statistical summary of the thee-way ANOVA testing the effect of the different factors: CLIMATE, SEASON and UV on litter decomposition rate (*k*).

Factor	**η^2**	**MS**	**DenDF**	**NumDF**	**F value**	***P***
SEASON	0.005	0.005	30	1	6.02	**< 0.05**
CLIMATE	0.105	0.122	30	1	140.73	**< 0.001**
UV	0.255	0.148	30	2	170.53	**< 0.001**
SEASON:CLIMATE	0.003	0.004	30	1	4.38	**< 0.05**
SEASON:UV	0.023	0.013	30	2	15.39	**< 0.001**
CLIMATE:UV	0.001	0.000	30	2	0.3	0.717
SEASON:CLIMATE:UV	0.000	0.000	30	2	0.333	0.720

Litter decomposition rates were higher in the CONTROL than in the WARMING treatment (Figure 6). Climate manipulation exerted a small to medium effect ($\eta^2 = 0.105$) on *k* ($F_{30,1} = 140.43$, $P < 0.001$), significantly lowering *k* compared to the control treatments (Figure 6). Differences between radiation treatments were consistent in the WARMING treatment with higher decomposition rates in the control than in the filter treatments, particularly in the DRY period (more than double). Differences diminished during the WET period after watering, but to a lesser extent than in the CONTROL treatment. Although differences between the UV treatments were not significant ($P < 0.05$), litter under the +UV treatment always had higher decomposition rates than in the -UV treatment (*ca*. 13%).

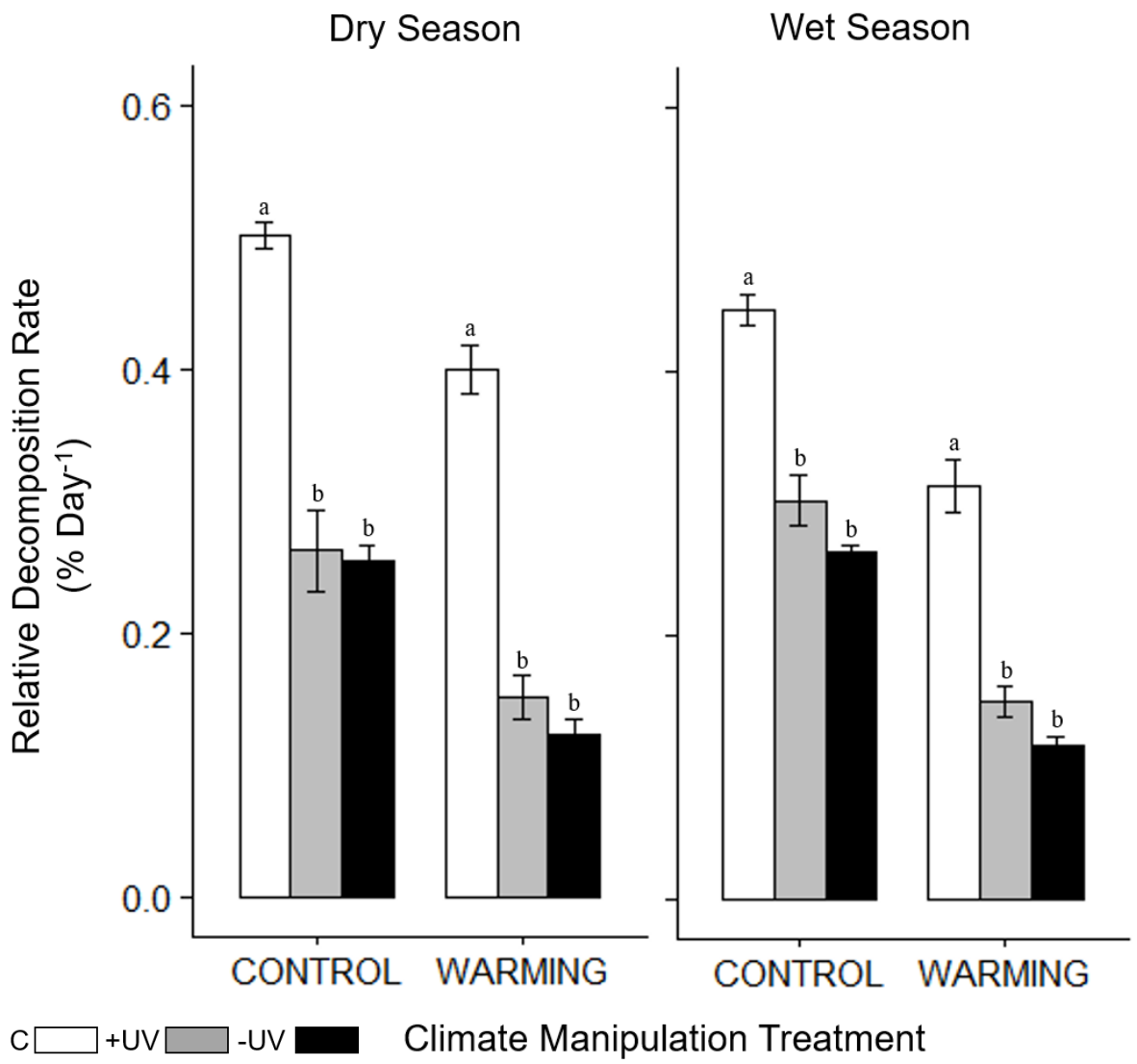

Figure 6. Relative decomposition rate *k* ($\%_{\text{mass loss}}$ ash free dry mass day^{-1}) over the DRY season and WET seasons in the climate manipulation treatments: CONTROL and WARMING for the three light manipulation treatments: control, -UV and +UV represented by white, grey and black bars, respectively. Bars represent the mean ± 1SE (n = 6). Bars with different letters are significantly different (P < 0.05), lower case letters represent differences within climate treatments.

When litter decomposition rate was standardised for temperature and humidity (S3) there was little difference between -UV and +UV treatments (2.97%) in the DRY season. The difference, however, increased throughout the WET season (11.82%). This was also evident when looking at the climate manipulation treatments, where *k* was 24.94% and 53.38% lower than those in the CONTROL treatment in the DRY and WET seasons, respectively.

Litter mass loss ranged from 11.71 to 50.69% throughout the DRY season and 24.61 to 70.67% by the end of the WET season depending on climate and UV manipulation (Table 4) with CONTROL treatments always showing the highest mass loss and -UV showing the lowest mass loss. Litter decomposition was 14% and 23% higher during the dry period than in the wet period in the CONTROL and the WARMING treatments, respectively (Table 4).

Table 4. Mean mass remaining (%) depending on treatment and season ± 1 SE.

Season	Climate Manipulation	UV Treatment	Mass Remaining (%)
DRY	CONTROL	Control	49.31 ± 1.67
		+UV	66.54 ± 6.64
		- UV	71.71 ± 4.04
	WARMING	Control	57.19 ± 5.53
		+UV	82.75 ± 4.30
		- UV	88.29 ± 7.05
WET	CONTROL	Control	29.33 ± 2.13
		+UV	42.71 ± 5.00
		- UV	49.12 ± 8.45
	WARMING	Control	43.91 ± 6.00
		+UV	69.64 ± 5.13
		- UV	75.39 ± 2.97

Based on the litter decomposition rates measured in the different treatments and as, during the dry season, mass loss in the UV- was mostly the result of thermal degradation (Gliksman *et al.*, 2017), the difference between UV+ and UV- can be attributed to photodegradation and the difference with the mass loss in the control can be attributed mainly to microbial degradation. Amost 34%, 10% and 56% of the mass loss over the dry period can be attributed to microbial, photo and thermal degradation respectively in the CONTROL treatment. Warming lead to an increase in microbial decomposition of 26%, in photodegradation by 3% and a decrease in the relative influence of thermal decomposition of 28%.

Litter structural changes

At the end of the experiment, both litter chemistry and structural components showed significant differences amongst experimental manipulations (Figure 7). Litter chemistry significantly differed ($P < 0.05$ in all cases) between seasons (Table 5). During the DRY season, UV had the strongest effect on loss of the soluble cell fraction (SCF) ($F_{2,18} = 185.92$, $P < 0.001$) which was the largest loss from the C fraction showing a 63.1 ± 3.2 % reduction in mass. Significantly more of the SCF was lost in the control (70.02 ± 2.07%) than -UV (51.16 ± 3.71%) and +UV (53.67 ± 3.61%) treatments. Litter under the control treatment had lost significantly more SCF and cellulose than litter under filters. This trend was the same in the WARMING treatment. However, litter under filters lost significantly less SCF than in the CONTROL treatment. Although not significant, litter in the +UV treatment had lost slightly more SCF than litter in the -UV treatment. Under WARMING, the effect of -UV was more pronounced, with significantly less losses in hemicellulose and cellulose. There was a small but significant increase in the SCF by the end of the WET season (7.0 ± 3.33%, $F_{1,18} = 19.039$, $P < 0.001$). Between the DRY and WET season there was a clear reduction in cellulose (35.82 ± 4.82%, $F_{1,18} = 711.5$, $P < 0.001$) and hemicellulose (58.66 ± 4.70%, $F_{1,18} = 571.26$, $P < 0.001$) content (Table 5). Cellulose, hemicellulose and SCF were significantly lower in wet season UV controlled compared to all forms of -UV and +UV (Figure 7). In the WET season, structural changes in the CONTROL treatment differed from those in the DRY season. Litter in the control treatment had lost more cellulose and lignin than the filter treatments while no differences in any component were detected between filter treatments. In the WARMING treatment, all structural components were higher than in the CONTROL treatment except for lignin. Warming led to a significant decrease in lignin like compounds (90.17 ± 8.75%, $F_{1,18} = 29.18$, $P < 0.001$) compared to the CONTROL-control (158.13 ± 16.54%). Differences between

+UV and -UV were only significant for the SCF, with a larger quantity remaining in the +UV than in the -UV ($P < 0.05$).

At the end of the DRY season, the LCI did not significantly differ among treatments (Table 5). However, after the WET season, LCI was significantly higher in the CONTROL treatment (0.37 ± 0.02) compared to the WARMING treatment (0.17 ± 0.02) with climate manipulation having a large effect on LCI ($F_{1,18} = 72.63$, $P < 0.001$). Climate manipulation had the strongest effect on ash content ($F_{1,23} = 52.12$, $P < 0.001$), being significantly higher in the DRY season control and +UV than in the WARMING treatments and significantly higher in all climate CONTROL compared to WARMING in the WET season (S2).

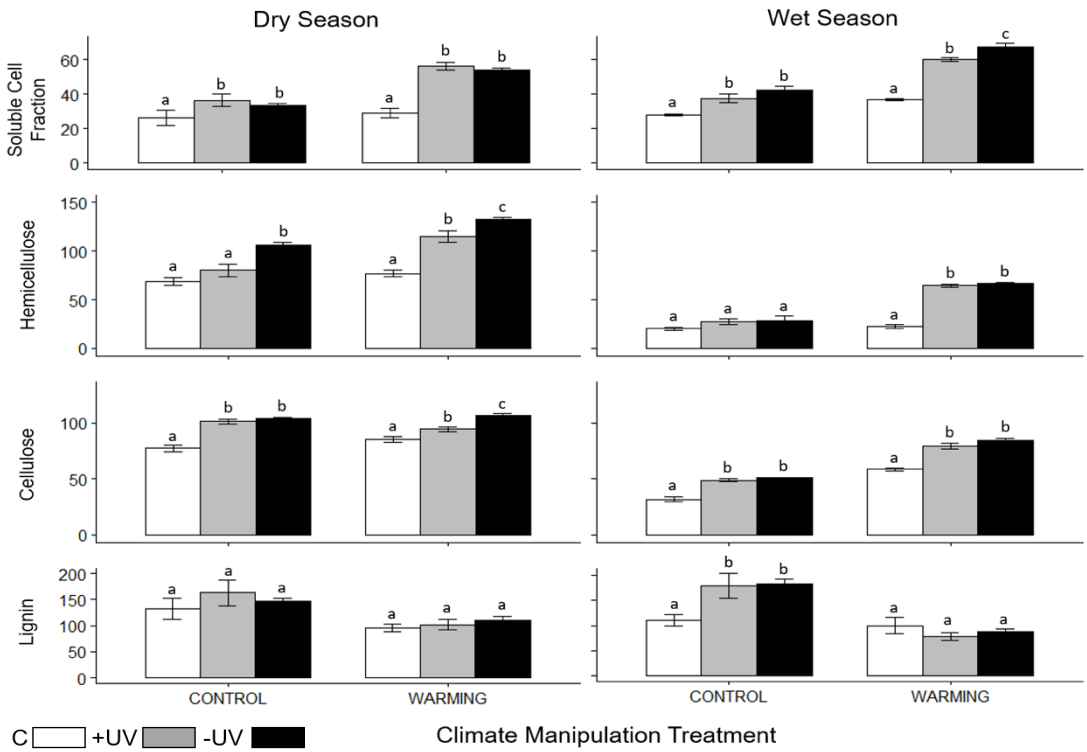

Figure 7. Main structural components of litter (% remaining nutrient) after the dry and wet periods in the climate manipulation treatments: CONTROL and WARMING for the three light manipulation treatments: control, -UV and +UV represented by white, grey and black bars respectively. Bars represent the mean ± 1SE ($n = 6$). Bars with different letters are significantly different ($P < 0.05$), lower case letters represent differences within climate treatments.

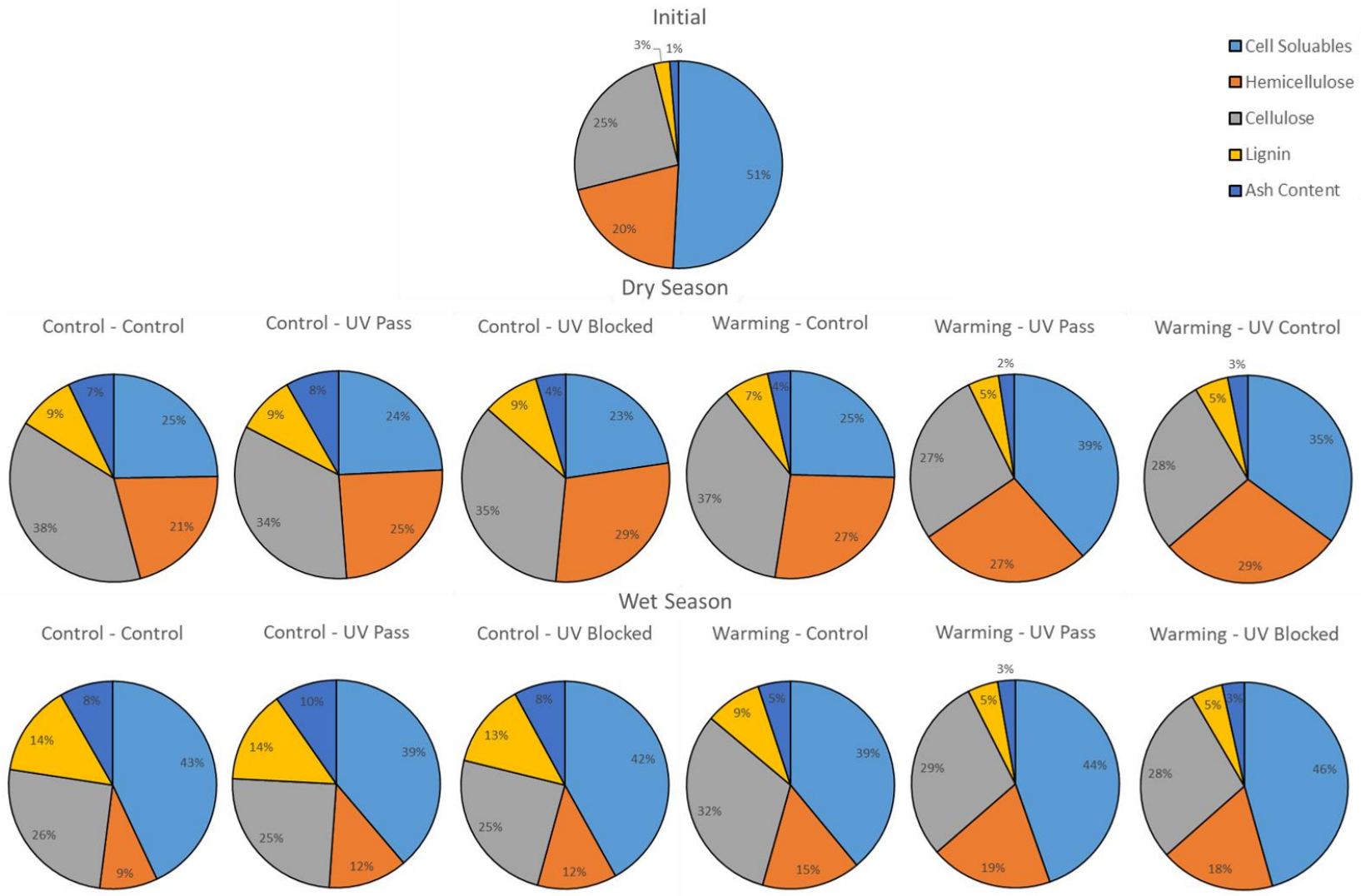

Figure 8. Treatment and season effect on chemical constituents.

The C:N ratio was unaffected by all types and combinations of climate manipulation and season. Season exerted the strongest effect upon water content ($F = 91.06$, $P < 0.001$) with a reduction from 4.03 ± 0.13% in the DRY season to 2.54 ± 0.10% in the WET season (Table 5). Litter moisture was not affected by either climate or radiation treatments, while the ash content was significantly different between climate manipulation treatments.

Table 5. Statistical results of the three-way ANOVA testing the effect of the different factors: CLIMATE, SEASON and UV on structural components.

Variable	Factor	η^2	Mean Sq	DF	F value	*P*
	SEASON	0.716	19.854	1	97.06	< **0.001**
Litter moisture	CLIMATE	0.032	0.883	1	4.32	0.057
	UV	0.023	0.324	2	1.58	0.240
	SEASON	0.054	27.121	1	7.66	< **0.05**
Ash content	CLIMATE	0.369	184.541	1	52.12	< **0.001**
	UV	0.022	5.397	2	1.524	0.242
	SEASON	0.159	1.131	1	26.10	< **0.001**
	CLIMATE	0.443	3.148	1	72.63	< **0.001**
	UV	0.014	0.050	2	1.14	0.341
LCI	SEASON:CLIMATE	0.124	0.879	1	20.27	< **0.001**
	SEASON:UV	0.015	0.055	2	1.27	0.306
	CLIMATE:UV	0.018	0.065	2	1.49	0.251
	SEASON:CLIMATE:UV	0.006	0.023	2	0.52	0.601
	SEASON	0.024	177.49	1	19.04	< **0.001**
	CLIMATE	0.248	1831.72	1	196.49	< **0.001**
	UV	0.470	1733.25	2	185.92	< **0.001**
Soluble cell fraction	SEASON:CLIMATE	0.001	8.710	1	0.93	0.347
	SEASON:UV	0.020	72.50	2	7.78	< **0.01**
	CLIMATE:UV	0.029	105.77	2	11.35	< **0.001**
	SEASON:CLIMATE:UV	0.002	7.299	2	0.78	0.472
	SEASON	0.576	15443.11	1	711.52	< **0.001**
	CLIMATE	0.113	3039.93	1	140.06	< **0.001**
	UV	0.176	2359.57	2	108.71	< **0.001**
Cellulose	SEASON:CLIMATE	0.094	2519.96	1	116.10	< **0.001**
	SEASON:UV	0.001	16.31	2	0.75	0.486
	CLIMATE:UV	0.004	47.93	2	2.21	0.139
	SEASON:CLIMATE:UV	0.006	83.03	2	3.83	< **0.05**
	SEASON	0.624	40715.77	1	571.26	< **0.001**
	CLIMATE	0.093	6096.23	1	85.53	< **0.001**
	UV	0.143	4672.03	2	65.55	< **0.001**
Hemicellulose	SEASON:CLIMATE	0.000	20.98	1	0.29	0.594
	SEASON:UV	0.017	540.22	2	7.58	< **0.05**
	CLIMATE:UV	0.029	944.70	2	13.26	< **0.001**
	SEASON:CLIMATE:UV	0.003	85.10	2	1.194	0.326
	SEASON	0.001	76.69	1	0.058	0.812
Lignin	CLIMATE	0.384	38738.97	1	29.18	< **0.001**
	UV	0.050	2541.49	2	1.91	0.174

Enzyme Activities

Throughout the duration of the study no oxidative enzyme activity was detected (data omitted). All hydrolytic enzymes assayed were significantly higher in the WET season than in the DRY season: phosphatase ($F_{1,23} = 6.589$, $P < 0.05$), xyloside ($F_{1,18} = 11.653$, $P < 0.001$) and N-Acetyl-Glucosaminide ($F_{1,18} = 5.998$, $P < 0.05$) with a large effect size ($\eta^2 > 0.5$) (Table 6, Figure 9).

Table 6. Statistical results of the three-way ANOVA testing the effect of the different factors: CLIMATE; SEASON and UV on enzymatic activities.

Enzyme	Factor	η^2	Mean Sq	DF	F value	*P*
Phosphatase	SEASON	0.567	8.840	1	68.92	< **0.001**
	CLIMATE	0.054	0.845	1	6.59	< **0.05**
	UV	0.021	0.166	2	1.29	0.297
Cellobiohydrolase	SEASON	0.725	764.48	1	557.16	< **0.001**
	CLIMATE	0.004	4.22	1	3.07	0.097
	UV	0.006	2.34	2	1.71	0.209
	SEASON:CLIMATE	0.062	48.74	1	35.52	< **0.001**
	SEASON:UV	0.048	16.28	2	11.86	< **0.001**
	CLIMATE:UV	0.005	2.97	2	2.16	0.147
	SEASON:CLIMATE:UV	0.007	1.95	2	1.42	0.267
Glucopyranoside	SEASON	0.539	48.89	1	139.25	< **0.001**
	CLIMATE	0.007	0.61	1	1.75	0.203
	UV	0.011	0.50	2	1.41	0.270
	SEASON:CLIMATE	0.096	8.68	1	24.71	< **0.001**
	SEASON:UV	0.059	2.67	2	7.59	< **0.01**
	CLIMATE:UV	0.008	0.34	2	0.98	0.394
	SEASON:CLIMATE:UV	0.025	1.12	2	3.18	0.066
Xyloside	SEASON	0.971	26.11	1	4199.52	< **0.001**
	CLIMATE	0.003	0.072	1	11.65	< **0.001**
	UV	0.001	0.015	2	2.45	0.115
	SEASON:CLIMATE	0.001	0.033	1	5.31	< **0.05**
	SEASON:UV	0.002	0.023	2	3.72	< **0.05**
	CLIMATE:UV	0.009	0.115	2	18.57	< **0.01**
	SEASON:CLIMATE:UV	0.005	0.073	2	11.76	< **0.001**
N-Acetyl-Glucosaminide	SEASON	0.634	305.74	1	86.68	< **0.001**
	CLIMATE	0.044	21.16	1	5.99	< **0.05**
	UV	0.008	1.87	2	0.53	0.597

UV light alone did not significantly affect any hydrolytic enzymes, however the effect of UV on cellobiohydrolase and glucopyranoside activity differed significantly between season ($P < 0.001$) while the interaction effect was small ($\eta^2 = 0.05$). In the WET season, cellobiohydrolase

and xyloside activities were significantly lower in the control treatment (107.67 ± 9.84, 77.11 ± 7.97 nmol h^{-1} g $AFDM^{-1}$) compared to -UV (133.67 ± 20.67, 48.33 ± 1.67 nmol h^{-1} g $AFDM^{-1}$, $P < 0.001$) and +UV (199.33 ± 22.32, 95.0 ± 16.17 nmol h^{-1} g AFDM-1, $P < 0.001$).

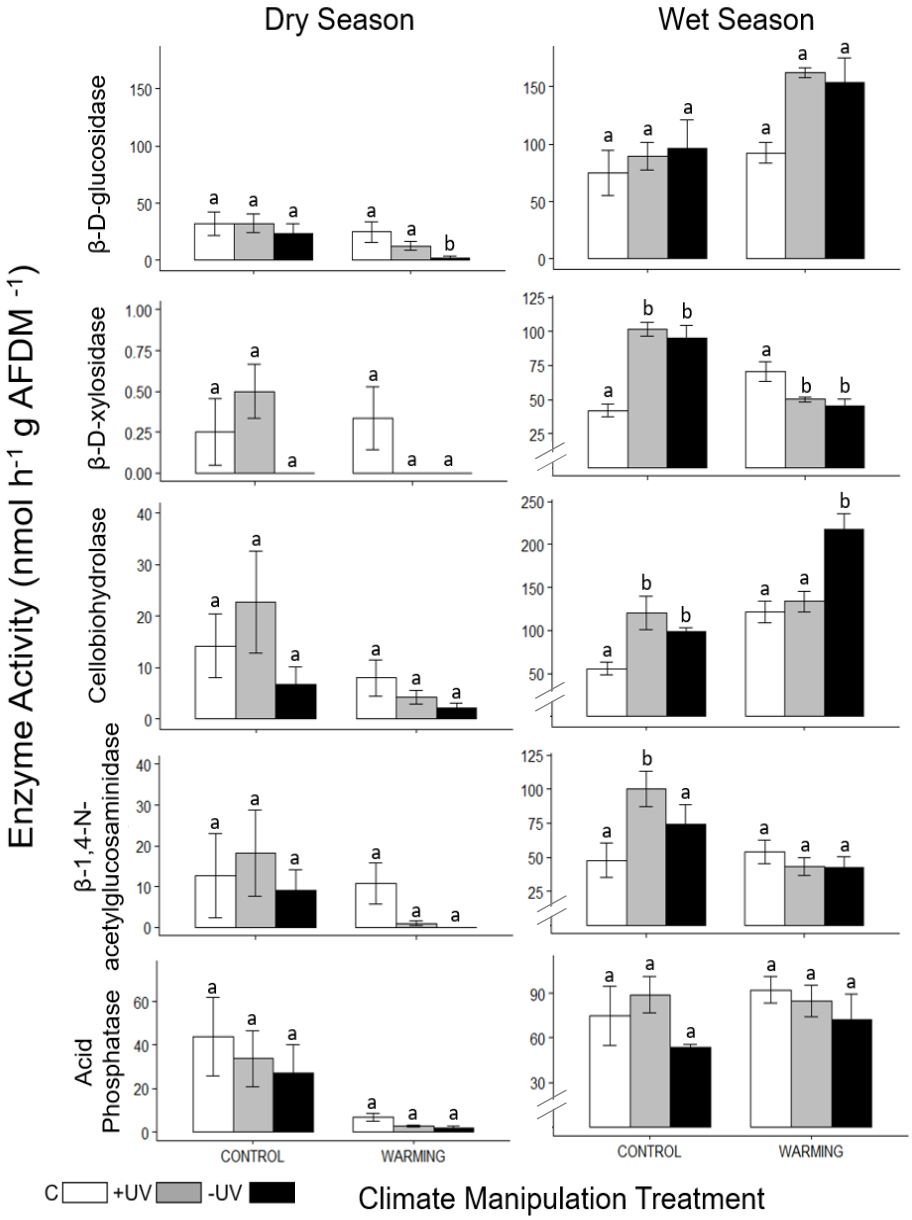

Figure 9. Enzymatic activities in the litter material (nmol h^{-1} g $AFDM^{-1}$) after the DRY season (A) and WET season (B) in the climate manipulation treatments: CONTROL and WARMING treatments for the three light manipulation treatments: control, -UV and +UV represented by white, grey and black bars respectively. Bars represent the mean ± 1SE ($n = 6$). Bars with different letters are significantly different ($P < 0.05$).

Discussion

By the end of the experiment, litter mass loss ranged from 21.4 to 73.4% depending on climate and UV manipulation which is higher than documented in most semi-arid grassland studies (e.g. Austin & Vivanco, 2006; Brandt *et al*., 2007; Austin & Ballare, 2010; Brandt *et al.*, 2010; Lin *et al*., 2015). Interestingly, the mass loss rates throughout the early stage of decomposition in this study far exceeds the best photodegradation model produced from the Long-term Intersite Decomposition Experiment Team data set produced by Adair *et al.*, (2017). This can be attributed to the selected litter mix being common and herbaceous, highly degradable with a low C:N ratio, low levels of lignin and a low lignin:cellulose index allowing us to detect a variety of effects of climate and UV manipulation over the short duration of the experiment (Figure 8). The data highlights the complexity of the controlling factors of dryland decomposition, how greatly they vary from non-water-limited systems and why current models, which do not account for such factors, systematically underestimate dryland decomposition.

Treatment manipulation effects

Using both filters and open top chambers to manipulate radiation levels and temperature caused significant changes in moisture dynamics due to the intrinsic relationship between temperature and relative humidity. Throughout both seasons there was a marked difference in air temperature and air relative humidity between climate manipulation treatments and a marginal difference between radiation treatments. Introducing UV filters decreased the transmission of visible and infra-red light, but also increased air temperature, blocked some wind and precipitation (Aphalo *et al.*, 2012). In this study, the effect of filters was small, less than 1^{o}C increase in temperature and little effect on precipitation since during the dry season hardly rained (52 mm in three events) and the filters had holes to allow water passage. The effect of

the filters on wind was expected to be small since the site is not windy and the filters were placed 20 cm above the soil surface. Therefore, the main effect of the filters was the amount of radiation and UV light that litter received, and the amount of water deposited overnight on the litterbags. Based on the reduction in air relative humidity below the 70% critical threshold to sustain microbial activity (Gliksman *et al*., 2017), these alterations led to a 4.02 ± 0.13% reduction in the total time that microbes could be potentially active throughout the dry season. However, this is probably highly underestimated, as water droplets fell on litter and the filters were often wet in the early morning indicating that the main effect was to prevent dew deposition on the ground (Rey, personal observation).

The use of open top chambers achieved the aimed average 2-3°C temperature increase in the warming treatment. This warming effect is mirrored by Maestre *et al.*, (2013) and Almagro *et al.*, (2015) using the same OTC design as this study. However, the temperature increase was often much greater at midday (up to ≈10°C) and less pronounce at night (up to ≈2°C). The OTCs also affect wind speed and inevitably influence the energy balance of soils due to calmer conditions reducing heat dispersion (De Boeck *et al*., 2012). As a result of these changes, particularly the slight increase in temperature, decrease in air relative humidity at night, and thus, reduction in dew formation, as observed also by Gliksman *et al*. (2017). The water deposited on the litter surface is enough to temporarily increase litter moisture content above the threshold value of 30% litter moisture content or 75% relative humidity determined by and thus, affected the time that microbes are active precluding litter decomposition during the dry period. It was visually observed water accumulating over the filters. This water would have fallen on the litter and increase litter moisture in the control treatment activating microbes at night in this treatment. Thus, air relative humidity may not properly reflect litter moisture which is the variable that most likely affects biotic degradation. Thus, the time that microbes are activated in the control plots was probably underestimated since dew formation was not

monitored. This together with the photopriming effect caused by photodegradation during the day may further accelerate litter decomposition and help explain the observed large differences in litter decomposition rates suggesting that both processes, biotic and abiotic processes were involved in litter decomposition.

Seasonal dynamics of litter decomposition

This study showed that the control of litter decomposition in water-limited systems is fundamentally different from those in wetter systems. Above all, it was found that as much litter decomposition takes place over the dry season (with practically no rainfall, high radiation and temperatures), as during the wet season. Traditional decomposition models suggest that the rate-limiting drivers of decomposition are abiotic (mostly temperature and moisture) and biotic (i.e. litter quality) which act to mediate decomposer community composition and metabolic activity (Vernon Meentemeyer, 1978; King *et al.*, 2012; Barnes *et al.*, 2015). In seasonal or dry ecosystems microbial activity is supressed by either low RH (<70%) (Gliksman *et al.*, 2017) or litter moisture contents below 30% (Prescott, 2010; Djukic *et al.*, 2018), and models typically underestimate decomposition rates by 30% (e.g. Whitford, 1981; Moorhead & Reynolds, 1991; Kemp *et al.*, 2003; Adair *et al.*, 2008; Throop & Archer, 2009). This study, in line with a growing body of literature (Austin & Vivanco, 2006; King *et al.*, 2012; Lee *et al.*, 2012; Austin *et al.*, 2016; Gliksman *et al.*, 2017; Lin *et al.*, 2018) revealed that non-traditional abiotic drivers (photochemical and thermal degradation) can have a disproportionally large effect on dryland litter decomposition, particularly thermal degradation which in this study contributed ca. 56% while photodegradation had a much smaller contribution (less than 13%) (H_1). In a meta-analysis conducted by King *et al.* (2012), studies testing the effect of photodegradative effects in moisture limited environments revealed that exposure to ambient UV increased mass loss by 32% on average. This stark increase in mass loss, in-line with this study, indicates that non-traditional drivers alone are not sufficient to

cause the extreme mass loss (50.52 ± 1.03%) during the short (four month) dry season in the open-air controls (H_4). Here, I suggest that litter moisture increases at night triggered microbial activity confirming previous studies (Dirks *et al.,* 2015) showing that relative humidity and possibly dew (Gliksman *et al*., 2017) are important water sources for microbes in these dry environments (H_1). Besides, interactions between photo, thermal, microbial decomposition and leaching probably contributed to dry season decomposition. Gliksman *et al*. (2017) identified a new mechanism of litter decomposition that operates at the diurnal scale. During the day, with high temperatures and radiation inputs and very low litter moisture content, microbial decomposition is absent and most of the degradation occurs as a result of photodegradation and thermal degradation, while at night, relative humidity and lower temperatures caused dew to increase litter moisture content enough to activate microbes. These processes, abiotic degradation at day, and biotic degradation at night, interact through a positive feedback, so that photo-thermally degraded litter decomposed faster by microbes, and microbial decomposition accelerates abiotic degradation. This study supports that magnitude of the effects of the various biotic, abiotic and physical processes occurring over the dry period are similar to rates measured over the wet season. It also suggests that the most meaningful controlling factor is moisture and failing to consider this effect could result in strong underestimation of litter decomposition.

Although the filters have holes drilled to allow rainfall to pass through, it is probable that the levels and intensity of rain reaching the litterbags were substantially reduced, as previously observed in other studies (Day *et al.,* 2018), minimising the effect of leaching. However, given that there were only four days of rain over the dry period and that all plots were watered during the wet period to avoid differences between treatments, this effect was probably very small. Mean air temperature and relative humidity in the open-air control treatments were 0.68 ± 0.05°C cooler and 1.64 ± 0.20% higher at night. Although these are a small variations,

combined with the passive warming effect of radiation filters (Gliksman *et al.*, 2018), they would significantly reduce dew deposition in the early hours of the morning, and reduce microbial activity in filtered treatments. Gliksman *et al.*, (2017, 2018) concluded that litter decomposition in the UV+ treatment was mostly the result of photodegradation with reduced microbial degradation and/or activity and the UV- treatment was mostly attributed to thermal degradation with reduced photochemical and microbial degradation.

In a laboratory study, Van Asperen *et al.* (2015) found that thermal degradation became a dominant abiotic decomposition process at temperatures above 25°C. Mean day time temperatures throughout the dry season in this study exceeded 30°C with maximum temperatures ranging from 56.1 to 61.9°C, implying that thermal degradation in this study is potentially of importance. Given the small differences in litter decomposition rates between UV+ and UV-, thermal degradation seems to be more important than photodegradation in this study accounting for up to 56% of the decomposition observed during the dry period (H_1).

Structural changes caused by litter decomposition processes

Interestingly, thermal degradation led to substantial decomposition of the soluble cell fraction (the most labile C) (59.9 ± 0.6% reduction). This contrasts with previous research which states that thermal degradation typically affects more recalcitrant compounds (Lee, Rahn & Throop, 2012). I believe that this is due to the ubiquitous presence of fungi throughout the treatments which break down litter through the secretion of extracellular enzymes and via physical fragmentation through the growth of hyphae. The two main species examined in this study, *F. arundinacea* and *L. perenne*, are known to have a symbiotic relationship with fungal endophytes when growing (Henning *et al.*, 2000). Promputtha *et al.*, (2010) found compelling evidence from the succession of extracellular enzymes produced by fungal endophytes in senesced leaves to support the theory that fungal endophytes become saprophytes post-

senescence, aiding decomposition. Although fungal colonization was not quantitatively assessed in this study, the activity of ß-1-4-N-acetlyglucosaminidase (NAG) (Figure 9), which was uniform and throughout all levels of UV in the control treatment at the end of the dry season, can be used as an indicator of fungal presence as it chiefly degrades chitin in fungal cell walls (Talbot & Treseder, 2012). In a study conducted by Barnard *et al.* (2013) communities at three Californian grassland sites were found to be unaffected by summer dry or autumn wet conditions, indicating their marked resistance to changes in water availability and ability to cope with drought. The uniformity of NAG activity suggests that fungi were present at similar levels for all UV treatments. In order for the endophytes to colonize during the early stages of litter decomposition they need a ready supply of soluble sugars (labile carbon) (Petrini, 1991; Petrini *et al.*, 1993) potentially explaining the high rates of labile carbon decomposition in UV-/+ treatments where reduced microbial activity is present. The additional, significant loss of labile carbon in the control treatments can, thus, be explained by the increase in microbial activity. This is further supported by the increase in NAG activity in the warming – control treatment displaying a significant reduction in labile carbon compared to the UV-/+ treatments.

Even though photodegradation contributed little to litter decomposition, it led to a significant reduction in hemicellulose content by 26.30%, but had only a small, non-significant effect on mass loss (3.69%). This reduction cannot, like labile carbon, be explained by microbial or fungal decomposition due to the near non-existent enzyme activity throughout the dry season. This result is in-line with multiple other studies (e.g. Rozema *et al.*, 1997; Brandt *et al.*, 2010; Baker & Allison, 2015; Lin *et al.*, 2015; Lin, Scarlett & King, 2015; Adair *et al.*, 2017; Huang & Li, 2017). In a multi-site, multispecies litter decomposition experiment, Brandt *et al.*, (2010) found reductions in hemicellulose due to photodegradation for both studies species at all three sites for all time-points except two. Hemicellulose functions as a structural component of the

primary and, when cross-linked with lignin, secondary cell walls whereby it encases cellulose (Chen, 2014). Photodegradation of lignin is a strongly debated topic in the literature with some studies finding no clear effects on the lignin fraction (Foereid *et al.*, 2010; Kirschbaum, Lambie & Zhou, 2011; Lin *et al.*, 2015, 2018; Adair *et al.*, 2017) and others finding higher lignin loss due to UV radiation (Austin and Ballare, 2010; Huang *et al.*, 2017). As hemicellulose is photodegraded, the structure of the lignified secondary cell wall can be assumed to weakened as the chemical bonds of the hemicellulose are cleaved by free radicals produced via indirect photolysis (Crutzen *et al*., 1999; Brandt *et al.*, 2010; Lin *et al.*, 2015). When mass loss exceeds 40%, degradation of lignified tissue has been found to ensue (Djukic *et al.*, 2018). Many other studies have found that photodegradation reduces lignin content (Day *et al*., 2007; Henry *et al*. 2008; Austin & Ballare, 2010; Song *et al.*, 2013; Lin *et al.*, 2015; Austin *et al*. 2016; Huang & Li, 2017). These results, from low-lignin litter (2.52 ± 0.07%), are supportive of Brandt *et al.* (2009) who found that photodegradation did not affect lignin decomposition, despite studying litter types with varying lignin contents. This is further supported by Lin *et al.*, (2015) who found that hemicellulose is more sensitive than lignin in some species. The degradation of hemicellulose and weakening of the secondary cell wall is supportive of the well documented photopriming mechanism (Day *et al*., 2007; Gallo *et al.*, 2009; Foereid *et al.*, 2010; Barnes *et al.*, 2015; Lin *et al.*, 2015, 2018; Wang *et al*., 2015; Austin *et al*., 2016; Gliksman *et al.*, 2017; 2018) (H_4). This mechanism is further supported by the marginal decrease in cellulose content (2.88%) and increase in labile carbon (8.97%) due to photodegradation. For cellulose to be broken down, the secondary cell wall must be weakened via either lignin or hemicellulose breakdown (Chen, 2014), exposing carbohydrates to microbial breakdown (Austin *et al*., 2016) which can be seen with the increase in labile carbon.

The significant increase in litter decomposition rate (0.24% d^{-1}), decrease in labile carbon (17.11%), cellulose (23.59%) and hemicellulose (19.65%) in the control compared to the

values observed in the +UV treatment is likely due to a combination of leaching and photo-microbial feedback. The exact quantification of each process is difficult as microbial biomass had to be omitted due to a mechanical error in the analysis. However, the literature and enzyme assays give a good indication as to which processes are most likely to account for the loss of the relative fractions. Microbial communities face a series of stressors in arid and semi-arid systems due limited access to water. To prevent complete desiccation and cell lysis, microbes must accumulate solutes to reduce their internal water potential (Harris, 1981) which is very energetically expensive (Schimel *et al., 2007)*. Due to the low rainfall throughout the dry season, microbes will not have had to expel solutes often as a result of re-wetting, and as such the excess labile carbon provided by photopriming could also act as a buffer to desiccation. It is also suggested that microbial community structure can adapt to become arid conditions, shifting to favour drought-tolerant species (Fierer *et al*., 2003; Schimel *et al*., 2007; Cotrufo *et al.*, 2013). The theoretical barrier of substrate availability constraining microbial activity in dry soils (Manzoni *et al*., 2012) may also be alleviated by the production of labile carbon and increased access to substrates through photodegradation. The low C:N should also supply more than enough organic N to sustain microbial activity during the dry season (Austin *et al.*, 2004). Although the enzyme assays revealed little activity at the end of the dry season, probably due to the relative air humidity and litter moisture content being below the threshold for microbial activity at the time of sampling, it is unlikely that microbial communities desiccated due to water-stress throughout the dry season as indicated by the presence of some hydrolytic enzyme activity (Figure 9).

The relative importance of the novel diurnal photopriming mechanism described by Gliksman *et al.*, (2017) has not been quantitively assessed in this study, but it could be crucially important. Throughout the dry season relative air humidity exceeded the threshold for microbial activity for about 25% of the study period, mostly in the early hours of the morning (H_1). Gliksman *et*

al., (2017) was, to my knowledge, the only other study to assess the effect of photodegradation using both: identical radiation filters and an open-air control. The authors attribute the increased mass loss (+50%) in their no-screen control to a combination of microbial decomposition and co-occurring priming processes. It is for these same reasons, I believe that increased loss in cellulose and hemicellulose was observed in the control treatment.

The increased loss of labile carbon in the control was likely due to leaching for three possible reasons; i) the increased access of soil microorganisms makes the leaching of labile carbon more likely (Gallo *et al.*, 2009), ii) the increased solubility of photodegraded litter leads to increased leaching (Gallo *et al.*, 2006; Feng *et al.*, 2011) and iii) the favourable microsite conditions would allow increased levels of microbial colonization. These results highlight the importance of photo and thermal degradation, microbial decomposition and leaching as independent processes, but also that their combination can have a disproportionate effect on the overall decomposition of easily degradable litter, supporting my initial hypothesis (H_1).

Dry season decomposition effects subsequent wet season decomposition

During the wet season, all treatments were artificially watered with distilled water in order to alleviate the water inhibition on microbial decomposition ensuring that all treatments received equal amount of water avoiding any unwanted artefacts created by the filters. This means that any differences in wet season decomposition were either due to a priming effect of dry season decomposition, and UV inhibition on microbial activity.

These results indicate, that, even with the reduction of dry season microbial decomposition in the filtered treatments there was a seasonal priming effect due to UV light exposure (H_4). When litter decomposition rate was standardised for temperature and relative humidity, there was no difference between +UV and -UV treatments during the dry season. There was, however, a marginal increase throughout the wet season which was probably due to the mechanisms

mentioned above. This result reinforces the hypothesis that the large amount of litter decomposition in the control treatment compared with the filtered treatments was partly the result of a positive feedback mechanism between photo and thermal degradation on microbial decomposition.

When microbial decomposition was present during the dry season as indicated in the open-air control treatment, there was also a relative increase in standardised litter decomposition rate however there were no other direct signs of seasonal photopriming. I do, however, suggest that the air humidity mediated photo-microbial decomposition loop sufficiently weakened the secondary cell wall in the open-air control to lead to a significantly faster litter decomposition rates compared to filtered treatments and that a higher priming effect on chemical constituents would have been apparent if substrate availability had not been a limiting factor (Sinsabaugh *et al.*, 2008).

Besides priming effects, dry season decomposition caused a shift in the fractions of the cell being decomposed. Due to the ubiquitous consumption of labile carbon throughout the dry season and the shift in dominant processes from abiotic to biotic, wet season decomposition preferentially degraded the more recalcitrant compounds such as holocellulose (H_2).

Mechanisms of decomposition in the wet season

Due to the artificial watering regime throughout the wet season, it can be assumed that the capacity for microbial activity should be uniform throughout the treatments unless there was an inhibitory effect of UV light or filtering on microbial activity. In addition to the diurnal photopriming throughout the dry season (Gliksman *et al.*, 2017; Gliksman *et al*., 2018) there is evidence for seasonal photopriming (Austin *et al*., 2016) but only when full microbial activity was present throughout the dry season (H_3). UV light inhibited microbial activity throughout the wet season as indicated by a reduction in total hydrolytic enzyme activity in the

+UV vs -UV treatments (54.42 nmol h^{-1} g $AFDM^{-1}$) and control (86.27 nmol h^{-1} g $AFDM^{-1}$) which supports previous studies (eg. Kurbanyan *et al.*, 2003; Fernández Zenoff *et al*., 2006; Lin, *et al*. 2015). Interestingly, the positive effect of photopriming overcame this inhibition indicated by the significantly higher mass loss and faster litter decomposition rate in the control treatment and the ubiquitous increase in the Ligninocellulose index in all CONTROL treatments (H_3). This suggests that the well-documented inhibitory effect of UV light on microbial DNA (Rohwer & Azam, 2000) and microbial communities (Hughes *et al*., 2003) is surpassed by the increased substrate availability (Austin *et al*., 2016; Lin *et al.*, 2018). This occurs up to the point where substrate availability (Almagro *et al.*, 2016) and substrate quality decrease enough (Swift *et al*., 1979) to become limiting factors. This is indicated by a control showing an additional 31.85 nmol h^{-1} g $AFDM^{-1}$ decrease in hydrolytic enzyme activity (which was significant in 3/5 of the assayed enzymes) and an increase in recalcitrance indicated by a significant increase in the C:N ratio compared to the +UV treatment.

Warming effect on litter decomposition

Introducing OTCs achieved the 2-3°C temperature increase aimed for, but inevitably led to further decreasing RH and litter moisture content, which is expected under a warmer climate. Due to this, all treatments were expected to have lower microbial activity, especially in plots which are filtered due to the compounding effects of introducing filters with OTCs.

In line with the only study which, to my knowledge, assesses the effect of warming on abiotic dryland decomposition using OTCs, warming significantly reduces litter decomposition (Almagro *et al.*, 2015). In the CONTROL treatment, litter decomposition rates were only significantly faster when not standardised for temperature and relative humidity, which is to be expected due to their status as traditional drivers of decomposition, reinforcing their importance in dryland systems (Vernon Meentemeyer, 1978; Adair *et al.*, 2008).

OTCs reduced mean dry season day- and night-time RH by 4.84, and 2.41% respectively. The OTCs have a compounding effect with filters causing a further reduction in air relative humidity (Almagro *et al.*, 2015), dew formation (Maestre *et al.*, 2013; Gliksman *et al.*, 2017; Gliksman *et al*., 2018) and consequently on litter moisture content (Almagro *et al.*, 2015) leading to a reduction in microbial activity. This is supported by the reduction in total hydrolytic enzyme activity (73.55 nmol h^{-1} g $AFDM^{-1}$) in the warming -UV compared to the control -UV (H_5). Contrary to my initial theory, the relative contribution of thermal decomposition was reduced in the warming treatment. Thermal decomposition in the warming treatments decreased litter decomposition and increased soluble fractions, hemicellulose and cellulose content by 0.13% AFDM d^{-1}, 17.90%, 25.89% and 2.95% respectively compared to thermal decomposition in the CONTROL treatments. These results reveal the importance of even small amounts of microbial and fungal mediated decomposition in the control--UV treatment, highlighting its importance as a driver of dry season decomposition.

In line with Almagro *et al.* (2015), despite the slower litter decomposition rates, warming favoured photodegradation due to the further suppression of microbial activity as a consequence of drier conditions (Brandt *et al*., 2007; Smith *et al.*, 2010) and the significant reduction in ash content which is indicative of a reduction in soil-litter mixing (due to reduced soil transmission via wind in the OTCs). This both increases the risk of microbial desiccation (Moorhead & Reynolds, 1991) and reduces the rate of UV refraction from soil particles, increasing the risk of inhibition of microbial activity due to UV light (Wang *et al.*, 2015a).

Throughout the dry season, photodegradation was responsible for a significant depletion in cellulose (12.36% AFDM d^{-1}) and a significantly smaller increase in hemicellulose content, falling in line with the mechanisms of photodecomposition explained earlier. Due to the aforementioned reductions in microbial activity due to filtering and warming, the effect of photo-microbial decomposition was also clearer and more exacerbated in the warming

treatment, with significant depletions of soluble cell fractions (26.57 % AFDM), hemicellulose (38.25 % AFDM), cellulose (9.21% AFDM and lignin (9.25% AFDM). This reduction eliminated any signs of seasonal photopriming on litter decomposition and all chemical constituents except hemicellulose in the control, highlighting the importance of ambient moisture content for litter decomposition in these regions (H_5).

As expected, there was a significant increase in microbial activity throughout the wet season, indicated by ubiquitous increase in hydrolytic enzyme activity and a marginal increase in soluble cell fractions. Mostly the patterns of decomposition mimic the control treatment with less material decomposing. It is, however, interesting that UV does not appear to inhibit hydrolytic enzyme activity, but instead enforces it. I believe that this is probably due to the alleviation of substrate limitation (due to lower decomposition rates) combined with the compounded negative effects of filtering and OTCs on microbial activity.

In this study I refrained from making mechanistic postulations about the lignin fraction of the litter mix due to the inaccuracies produced via acid digestion. Although apt for understanding digestibility and litter solubility I recommend more precise quantifications such as those used Mansfield *et al.*, (2012) are used when assessing mechanisms.

The objective of this study was to investigate the processes involved in seasonal litter decomposition and the possible feedback mechanisms between abiotic and biotic processes of litter decomposition in order to better understand the potential mechanisms which account for the underestimation of carbon budgets in drylands. From this work combined with the suite of previous studies, it becomes clear that dry season litter decomposition is crucially important in water-limited systems. There is a complex array of mechanisms underlying the process which vary greatly depending on initial litter quality and climatic variability which fall into three categories: biotic, abiotic and physical processes. Previous literature has identified the relative importance of these processes individually but here I find that it is the interactions between

these processes that lead to the unexpectedly high decomposition rates over the dry season. Moreover, further evidence is provided to support the hypothesis that the whole process is mediated by night-time atmospheric moisture and water adsorption via dew. These findings highlight the importance of using open-air controls alongside UV filtering, without which studies risk failure to elucidate decomposition in dry systems properly, potentially missing out on the crucial diurnal feedback mechanisms that prove to be fundamental to this process. Further work is needed to untangle the complex interactions underlying litter decomposition in drylands. I believe that future research should be directed towards understanding the influence of photodegradation and night-time moisture on microbial and fungal community composition and function, investigating their effect on cell wall structural composition using 2DNMR based techniques. This would both allow us to better understand the potential mechanisms which account for the underestimation of carbon budgets in drylands and enable us to understand how climate change may impact forest carbon budgets as ecosystems continue to become warmer and drier.

References

Adair EC, Parton WJ, Del Grosso SJ, Silver WL, Harmon ME, Hall SA, Burke IC, Hart SC. **2008**. Simple three-pool model accurately describes patterns of long-term litter decomposition in diverse climates. *Global Change Biology* **14, pp. 2636–2660**.

Adair EC, Parton WJ, King JY, Brandt LA, Lin Y. **2017**. Accounting for photodegradation dramatically improves prediction of carbon losses in dryland systems. *Ecosphere* **8**.

Aerts R. **1997**. Climate, Leaf Litter Chemistry and Leaf Litter Decomposition in Terrestrial Ecosystems: A Triangular Relationship. *Oikos* **79, p. 439**.

Aerts R. **2006**. The freezer defrosting: Global warming and litter decomposition rates in cold biomes. *Journal of Ecology* **94, pp. 713–724**.

Almagro M, Maestre FT, Martínez-López J, Valencia E, Rey A. **2015**. Climate change may reduce litter decomposition while enhancing the contribution of photodegradation in dry perennial Mediterranean grasslands. *Soil Biology and Biochemistry* **90, pp. 214–223**.

Almagro M, Martínez-López J, Maestre FT, Rey A. **2016**. The Contribution of Photodegradation to Litter Decomposition in Semiarid Mediterranean Grasslands Depends on its Interaction with Local Humidity Conditions, Litter Quality and Position. *Ecosystems* **20, pp. 527–542**.

Andrews JA, Schlesinger WH. **2000**. Soil Respiration and the Global Carbon Cycle. *Biogeochemistry* **48, pp. 7–20**.

Anesio AM, Denward CMT, Tranvik LJ, Granéli W. **1999**. Decreased bacterial growth on vascular plant detritus due to photochemical modification. *Aquatic Microbial Ecology* **17**.

Ankom Technology. **2016**. *Determining Acid Detergent Lignin*. *https://www.ankom.com/sites/default/files/document-files/Method_13_ADL_Method_A2000_RevE_4_10_15.pdf*

Ankom Technology. **2017a**. *Determining Acid Detergent Fiber in Feeds Filter Bag Technique*. *https://www.ankom.com/sites/default/files/document-files/Method_13_ADF_Method_A2000_RevE_4_10_15.pdf*

Ankom Technology. **2017b**. *Determining Neutral detergent fiber in feeds - filter bag technique (for A2000 and A2000I). https://www.ankom.com/sites/default/files/document-files/Method_13_NDF_Method_A2000_RevE_4_10_15.pdf*

Aphalo PJ, Albert A, Mcleod AR, Robson TM, Rosenqvist E. **2012**. *Beyond the visible: A handbook of best practice in plant UV photobiology.* Helsinki: COST Action FA0906 UV4growth.

Van Asperen H, Warneke T, Sabbatini S, Nicolini G, Papale D, Notholt J. **2015**. The role of photo- and thermal degradation for CO2 and CO fluxes in an arid ecosystem. *Biogeosciences* **12, pp. 4161–4174**.

Austin AT, Ballare CL. **2010**. Dual role of lignin in plant litter decomposition in terrestrial ecosystems. *Proceedings of the National Academy of Sciences* **107, pp. 4618–4622**.

Austin AT, Méndez MS, Ballaré CL. **2016**. Photodegradation alleviates the lignin bottleneck for carbon turnover in terrestrial ecosystems. *Proceedings of the National Academy of Sciences* **113, pp. 4392–4397**.

Austin AT, Vivanco LL. **2006**. Plant litter decomposition in a semi-arid ecosystem controlled by photodegradation. *Nature* **442, pp. 555–558**.

Austin AT, Yahdjian L, Stark JM, Belnap J, Porporato A, Norton U, Ravetta DA, Schaeffer SM. **2004**. Water pulses and biogeochemical cycles in arid and semiarid ecosystems. *Oecologia* **141, pp. 221–235**.

Baker NR, Allison SD. **2015**. Ultraviolet photodegradation facilitates microbial litter decomposition in a Mediterranean climate. *Ecology* **28, pp. 63–86**.

Barnard RL, Osborne CA, Firestone MK. **2013**. Responses of soil bacterial and fungal communities to extreme desiccation and rewetting. *ISME* **7, pp. 2229–2241**.

Barnes PW, Throop HL, Archer SR, Breshears DD, Mcculley RL, Tobler MA. **2015**. Sunlight and Soil–Litter Mixing: Drivers of Litter Decomposition in Drylands. *Progress in Botany* **76, pp. 273-302.**

B**erg, B., and C. McClaugherty. 1980.** Decomposition, humus formation, carbon sequestration. *Plant litter. 2nd ed. Berlin Heidelberg: Springer,* 2008.

Berg B., Staaf H. 1980. Decomposition Rate and Chemical Changes of Scots Pine Needle Litter . II . Influence of Chemical Composition. *Ecological Bulletins* **32, pp. 373–390**.

De Boeck HJ, De Groote T, Nijs I. **2012**. Leaf temperatures in glasshouses and open-top chambers. *New Phytologist* **194, pp. 1155–1164**.

Bonan GB, Hartman MD, Parton WJ, Wieder WR. **2013**. Evaluating litter decomposition in earth system models with long-term litterbag experiments: An example using the Community Land Model version 4 (CLM4). *Global Change Biology* **19, pp. 957–974**.

Brandt LA, Bonnet C, King JY. **2009**. Photochemically induced carbon dioxide production as a mechanism for carbon loss from plant litter in arid ecosystems. *Journal of Geophysical Research: Biogeosciences* **114, pp. 1–13**.

Brandt LA, King JY, Hobbie SE, Milchunas DG, Sinsabaugh RL. **2010**. The role of photodegradation in surface litter decomposition across a grassland ecosystem precipitation gradient. *Ecosystems* **13, pp. 765–781**.

Brandt LA, King JY, Milchunas DG. **2007**. Effects of ultraviolet radiation on litter decomposition depend on precipitation and litter chemistry in a shortgrass steppe ecosystem. *Global Change Biology* **13, pp. 2193–2205**.

Budel B, Weber B, Kuhl M, Pfanz H, Sultemeyer D, Wessels D. **2004**. Reshaping of sandstone surfaces by cryptoendolithic cyanobacteria: bioalkalization causes chemical weathering in arid landscapes. *Geobiology* **12, pp. 261-268.**

Chen H. **2014**. Biotechnology of lignocellulose: Theory and practice. *Springer, 2016.*

Chen F, Dixon RA. **2007**. Lignin modification improves fermentable sugar yields for biofuel production. *Nature Biotechnology* **23, pp. 759**.

Corbeels M. **2001**. Plant Litter and Decomposition: General Concepts and Model Approaches. *NEE Workshop Proceedings* **8, pp. 123.**

Cornwell WK, Cornelissen JHC, Amatangelo K, Dorrepaal E, Eviner VT, Godoy O, Hobbie SE, Hoorens B, Kurokawa H, Pérez-Harguindeguy N, *et al.* 2008. Plant species traits are the predominant control on litter decomposition rates within biomes worldwide. *Ecology Letters* **11, pp. 1065–1071**.

Cotrufo MF, Wallenstein MD, Boot CM, Denef K, Paul E. **2013**. The Microbial Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: Do labile plant inputs form stable soil organic matter? *Global Change Biology* **19, pp. 988– 995**.

Couteaux MM, Bottner P, Berg B. **1995**. Litter decomposition climate and litter quality. *Trends in Ecology and Evolution* **10, pp. 63–66**.

Crutzen PJ, Schade GW, Hofmann R-M. **1999**. CO emissions from degrading plant matter. *Tellus B* **51, pp. 889–908**.

Day TA, Bliss MS, Tomes AR, Ruhland CT, Guénon R. **2018**. Desert leaf litter decay: Coupling of microbial respiration, water-soluble fractions and photodegradation. *Global Change Biology* **24, pp. 5454–5470**.

Day TA, Zhang ET, Ruhland CT. **2007**. Exposure to solar UV-B radiation accelerates mass and lignin loss of Larrea tridentata litter in the Sonoran Desert. *Plant Ecology* **193, pp. 185–194**.

Denman KL, Hauglustaine D. **2007**. Couplings Between Changes in the Climate System and Biogeochemistry. No. LBNL-464E. Lawrence Berkeley National Lab (LBNL), Berkeley, CA (United States)

Djukic I, Kepfer-Rojas S, Schmidt IK, Larsen KS, Beier C, Berg B, Verheyen K, Caliman A, Paquette A, Gutiérrez-Girón A, *et al.* 2018. Early stage litter decomposition across biomes. *Science of the Total Environment* **42, 628**–**629**.

Feng X, Hills KM, Simpson AJ, Whalen JK, Simpson MJ. **2011**. The role of biodegradation and photo-oxidation in the transformation of terrigenous organic matter. *Organic Geochemistry* **42, pp. 262–274**.

Fernández Zenoff V, Siñeriz F, Farías ME. **2006**. Diverse responses to UV-B radiation and repair mechanisms of bacteria isolated from high-altitude aquatic environments. *Applied and Environmental Microbiology* **72, pp. 7857-7863**.

Fierer N, Schimel JP, Holden PA. **2003**. Influence of drying-rewetting frequency on soil bacterial community structure. *Microbial Ecology* **45, pp. 63-81**.

Foereid B, Bellarby J, Meier-Augenstein W, Kemp H. **2010**. Does light exposure make plant litter more degradable? *Plant and Soil* **333, pp. 275–285**.

Gallo ME, Porras-Alfaro A, Odenbach KJ, Sinsabaugh RL. **2009**. Photoacceleration of plant litter decomposition in an arid environment. *Soil Biology and Biochemistry* **41, pp. 1433–1441**.

Gallo ME, Sinsabaugh RL, Cabaniss SE. **2006**. The role of ultraviolet radiation in litter decomposition in arid ecosystems. *Applied Soil Ecology* **34, pp. 82–91**.

García-Palacios P, Maestre FT, Kattge J, Wall DH. **2013**. Climate and litter quality differently modulate the effects of soil fauna on litter decomposition across biomes. *Ecology Letters* **16, pp. 1045–1053**.

Gavazov KS. **2010**. Dynamics of alpine plant litter decomposition in a changing climate. *Plant and Soil* **337, pp.19-32**.

Gaxiola A, Armesto JJ. **2015**. Understanding litter decomposition in semiarid ecosystems: linking leaf traits, UV exposure and rainfall variability. *Frontiers in Plant Science* **6, pp. 1–9**.

German DP, Weintraub MN, Grandy AS, Lauber CL, Rinkes ZL, Allison SD. **2011**. Optimization of hydrolytic and oxidative enzyme methods for ecosystem studies. *Soil Biology and Biochemistry* **43, pp. 1387–1397**.

Giorgi F, Lionello P. **2008**. Climate change projections for the Mediterranean region. *Global and Planetary Change* **63, pp. 90-104**.

Gliksman D, Haenel S, Grünzweig JM. **2018**. Biotic and abiotic modifications of leaf litter during dry periods affect litter mass loss and nitrogen loss during wet periods. *Functional Ecology* **32, pp. 831–839**.

Gliksman D, Rey A, Seligmann R, Dumbur R, Sperling O. **2017**. Biotic degradation at night, abiotic degradation at day : positive feedbacks on litter decomposition in drylands. *Global Change Biology* **233, pp. 1564–1574**.

Gressel J. **2008**. Transgenics are imperative for biofuel crops. *Plant Science* **10, 246-263**.

Harris RF. **1981**. Effect of water potential on microbial growth and activity. *American Society of Agronomy* **1, 23–95**.

Henning J, Lacefield G, Johnson K, Ditsch D. **2000**. Forage Identification and Use Guide Forage Identification and Use Guide. *Kentucky Department of Agriculture*.

Henry HAL, Brizgys K, Field CB. **2008**. Litter decomposition in a California annual grassland: Interactions between photodegradation and litter layer thickness. *Ecosystems* **11, pp. 545–554**.

Huang G, Li Y. **2017**. Photodegradation effects are related to precipitation amount, precipitation frequency and litter traits in a desert ecosystem. *Soil Biology and Biochemistry* **115, pp. 383–392**.

Huang G, Zhao H mei, Li Y. **2017**. Litter decomposition in hyper-arid deserts: Photodegradation is still important. *Science of the Total Environment* **601**–**602, pp. 784-792**.

Hughes KA, Lawley B, Newsham KK. **2003**. Solar UV-B radiation inhibits the growth of Antarctic terrestrial fungi. *Applied and Environmental Microbiology* **69, pp. 1488–1491**.

Incerti G, Bonanomi G, Giannino F, Rutigliano FA, Piermatteo D, Castaldi S, De Marco A, Fierro A, Fioretto A, Maggi O, *et al.* 2011. Litter decomposition in Mediterranean ecosystems: Modelling the controlling role of climatic conditions and litter quality. *Applied Soil Ecology* **49, pp. 148–157**.

Stocker TF, Qin D, Plattner GK, Tignor M, Allen SK, Boschung J, Nauels A, Midgley, PM, *et al.* 2013. Climate Change 2013 - The Physical Science Basis. *Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change*.

Jackson CR, Tyler HL, Millar JJ. **2013**. Determination of Microbial Extracellular Enzyme Activity in Waters, Soils, and Sediments using High Throughput Microplate Assays. *Journal of Visualized Experiments* **80, pp. 1–9**.

Kemp PR, Reynolds JF, Virginia RA, Whitford WG. **2003**. Decomposition of leaf and root litter of Chihuahuan Desert shrubs: Effects of three years of summer drought. *Journal of Arid Environments* **21, pp. 21-39**.

King JY, Brandt LA, Adair EC. **2012**. Shedding light on plant litter decomposition: Advances, implications and new directions in understanding the role of photodegradation. *Biogeochemistry* **111, pp. 57–81**.

Kirschbaum MUF, Lambie SM, Zhou H. **2011**. No UV enhancement of litter decomposition observed on dry samples under controlled laboratory conditions. *Soil Biology and Biochemistry* **43, pp. 1300–1307**.

Kottek M, Grieser J, Beck C, Rudolf B, Rubel F. **2006**. World map of the Köppen-Geiger climate classification updated. *Meteorologische Zeitschrift* **15, pp. 259–263**.

Kurbanyan K, Nguyen KL, To P, Rivas E V., Lueras AMK, Kosinski C, Steryo M, González A, Mah DA, Stemp EDA. **2003**. DNA-protein cross-linking via guanine oxidation: Dependence upon protein and photosensitizer. *Biochemistry* **45, pp. 10269-10281**.

Lee H, Rahn T, Throop H. **2012**. An accounting of C-based trace gas release during abiotic plant litter degradation. *Global Change Biology* **18, pp. 1185–1195**.

Lin Y, Karlen SD, Ralph J, King JY. **2018**. Short-term facilitation of microbial litter decomposition by ultraviolet radiation. *Science of the Total Environment* **615, pp. 838–848**.

Lin Y, King JY, Karlen SD, Ralph J. **2015a**. Using 2D NMR spectroscopy to assess effects of UV radiation on cell wall chemistry during litter decomposition. *Biogeochemistry* **125, pp. 427–436**.

Lin Y, Scarlett RD, King JY. **2015b**. Effects of UV photodegradation on subsequent microbial decomposition of Bromus diandrus litter. *Plant and Soil* **395, pp. 263–271**.

Maestre FT, Escolar C, de Guevara ML, Quero JL, Lázaro R, Delgado-Baquerizo M, Ochoa V, Berdugo M, Gozalo B, Gallardo A. **2013**. Changes in biocrust cover drive carbon cycle responses to climate change in drylands. *Global Change Biology* **19, pp. 3835–3847**.

Mansfield SD, Kim H, Lu F, Ralph J. **2012**. Whole plant cell wall characterization using solution-state 2D NMR. *Nature Protocols* **7, pp. 1579–1589**.

Manzoni S, Schimel JP, Porporato A. **2012**. Responses of soil microbial communities to water stress: Results from a meta-analysis. *Ecology* **93, pp. 930–938**.

Mayer LM, Thornton KR, Schick LL, Jastrow JD, Harden JW. **2012**. Photodissolution of soil organic matter. *Geoderma* **170, pp. 314-321**.

McCalley CK, Sparks JP. **2009**. Abiotic gas formation drives nitrogen loss from a desert ecosystem. *Science* **326, pp. 837-840**.

Moorhead DL, Callaghan T. **1994**. Effects of increasing ultraviolet B radiation on decomposition and soil organic matter dynamics: a synthesis and modelling study. *Biology and Fertility of Soils* **18, pp. 19–26**.

Moorhead DL, Reynolds JF. **1991**. A general model of litter decomposition in the northern Chihuahuan Desert. *Ecological Modelling* **53, pp. 197-219**.

Pancotto VA, Sala OE, Cabello M, López NI, Robson TM, Ballaré CL, Caldwell MM, Scopel AL. **2003**. Solar UV-B decreases decomposition in herbaceous plant litter in Tierra

del Fuego, Argentina: Potential role of an altered decomposer community. *Global Change Biology* **9, pp. 1465–1474**.

Pancotto VA, Sala OE, Robson TM, Caldwell MM, Scopel AL, Aires B. **2005**. Direct and indirect effects of solar ultraviolet-B radiation on long-term decomposition. *Global Change Biology* **11, pp. 1982–1989**.

Parton W, Silver WL, Burke IC, Grassens L, Harmon ME, Currie WS, King JY, Adair EC, Brandt LA, Hart SC, *et al.* 2007. Global-Scale Similarities in Nitrogen Release Patterns During Long-Term Decomposition. *Science* **315, pp. 361–364**.

Paudel E, Dossa GGO, Blécourt M de, Beckschäfer P, Xu J, Harrison RD. **2015**. Quantifying the factors affecting leaf litter decomposition across a tropical forest disturbance gradient. *Ecosphere* **6, pp. 1–20**.

Petrini O. **1991**. Fungal Endophytes of Tree Leaves. In: Andrews J.H., Hirano S.S. (eds) Microbial Ecology of Leaves. Brock/Springer Series in Contemporary Bioscience. Springer, New York, NY **179-197**

Petrini O, Sieber TN, Toti L, Viret O. **1993**. Ecology, metabolite production, and substrate utilization in endophytic fungi. *Natural Toxins*. **1, pp. 185-196**

Pointing SB, Belnap J. **2016**. Microbial colonization and controls in dryland systems. *Nature Reviews* **10, pp. 551–555**.

Prescott CE. **2010**. Litter decomposition: What controls it and how can we alter it to sequester more carbon in forest soils? *Biogeochemistry* **1-3, 133-149**.

Promputtha I, Hyde KD, McKenzie EHC, Peberdy JF, Lumyong S. **2010**. Can leaf degrading enzymes provide evidence that endophytic fungi becoming saprobes? *Fungal Diversity* **41, pp. 89–99**.

Raich JW, Schlesinger WH. **1992**. The global carbon dioxide flux in soil respiration and its relationship to vegetation and climate. *Tellus B* **44, pp. 81-99**.

Rohwer F, Azam F. **2000**. Detection of DNA damage in prokaryotes by terminal deoxyribonucleotide transferase-mediated dUTP nick end labeling. *Applied and Environmental Microbiology* **66, pp. 1001–1006**.

Rozema J, Tosserams M, Nelissen HJM, Heerwaarden L Van, Broekman R a, Flierman N. **1997**. Stratospheric ozone reduction and ecosystem processes: enhanced UV-B radiation affects chemical quality and decomposition of leaves of the dune grassland species Calamagrostis epigeios. *Plant Ecology* **128, pp. 285–294**.

Rutledge S, Campbell DI, Baldocchi D, Schipper LA. **2010**. Photodegradation leads to increased carbon dioxide losses from terrestrial organic matter. *Global Change Biology* **16, pp. 3065–3074**.

Safriel U, Adeel Z, Puigdefabregas J, Lal R. **2005**. Dryland Systems. *Ecosystems and Human Well-Being: Current State and Trends*. **pp. 623–662**.

Saiya-cork KR, Sinsabaugh RL, Zak DR. **2002**. The effects of long term nitrogen deposition on extracellular enzyme activity in an Acer saccharum forest soil. *Soil Biology and Biochemistry* **34, pp. 1309–1315**.

Sala OE, Chapin FS, Armesto JJ, Berlow E, Bloomfield J, Dirzo R, Huber-Sanwald E,

Huenneke LF, Jackson RB, Kinzig A, *et al.* 2000. Global biodiversity scenarios for the year 2100. *Science* **287, pp. 1770-1774**.

Schimel J, Balser T, Wallenstein M. **2007**. Microbial Stress-Response Physiology and it's Implications for Ecosystem Functioning. *New Directions in Microbial Ecology* **88, pp. 1386–1394**.

Schipper LA, Hobbs JK, Rutledge S, Arcus VL. **2014**. Thermodynamic theory explains the temperature optima of soil microbial processes and high Q10 values at low temperatures. *Global Change Biology* **20, pp. 3578–3586**.

Schröter D, Cramer W, Leemans R, Prentice IC, Araújo MB, Arnell NW, Bondeau A, Bugmann H, Carter TR, Gracia CA, *et al.* 2005. Ecology: Ecosystem service supply and vulnerability to global change in Europe. *Science* **310, pp. 1333-1337**.

Sinsabaugh RL, Lauber CL, Weintraub MN, Ahmed B, Allison SD, Crenshaw C, Contosta AR, Cusack D, Frey S, Gallo ME, *et al.* 2008. Stoichiometry of soil enzyme activity at global scale. *Ecology Letters* **11, pp. 1252–1264**.

Smith WK, Gao W, Steltzer H, Wallenstein MD, Tree R. **2010**. Moisture availability influences the effect of ultraviolet-B radiation on leaf litter decomposition. *Global Change Biology* **16, pp. 484–495**.

Song X, Peng C, Jiang H, Zhu Q, Wang W. **2013**. Direct and Indirect Effects of UV-B Exposure on Litter Decomposition: A Meta-Analysis. *PLoS ONE* **8, pp. 1–9**.

Swift MJ, Heal OW, Anderson JM. **1979**. Decomposition in terrestrial ecosystems. *Review Literature And Arts Of The Americas* **5, pp. 12–24**.

Talbot JM, Treseder KK. **2012**. Interactions between lignin, cellulose, and nitrogem drive litter chemistry - decay relationships. *Ecology* **93, pp. 345–354**.

Throop HL, Archer SR. **2009**. Resolving the Dryland Decomposition Conundrum: Some New Perspectives on Potential Drivers. *Progress in Botany* **70, pp. 171–194**.

Verhoef HA, Verspagen JMH, Zoomer HR, Verhoef HA, Verspagen JMH, Zoomer HR. **2000**. Direct and indirect effects of ultraviolet-B radiation on soil biota, decomposition and nutrient fluxes in dune grassland soil systems. *Biol Fertil Soils* **31, pp. 366–371**.

Vernon Meentemeyer. **1978**. Macroclimate the Lignin Control of Litter Decomposition Rates. *Ecology* **59, pp. 465–472**.

Wang J, Liu L, Wang X, Chen Y. **2015a**. The interaction between abiotic photodegradation and microbial decomposition under ultraviolet radiation. *Global Change Biology* **21, pp. 2095–2104**.

Wang H, Richardson CJ, Ho M. **2015b**. Dual controls on carbon loss during drought in peatlands. *Nature Climate Change* **5, pp. 584–588**.

Wang Q, Wang S, Huang Y. **2009**. Leaf litter decomposition in the pure and mixed plantations of Cunninghamia lanceolata and Michelia macclurei in subtropical China. *Biology and Fertility of Soils* **45, pp. 371–377**.

Wardle DA, Bardgett RD, Klironomos JN, Seta H, Putten WH van der, Wall DH. **2004**. Ecological Linkages Between Aboveground and Belowground Biota. *Science* **304, pp. 1629–1633**.

Warren-Rhodes KA, Rhodes KL, Boyle LN, Pointing SB, Chen Y, Liu S, Zhuo P, McKay CP. **2007**. Cyanobacterial ecology across environmental gradients and spatial scales in China's hot and cold deserts. *FEMS Microbiology Ecology* **3, pp. 470-482**.

Whelan ME, Rhew RC. **2015**. Carbonyl sulfide produced by abiotic thermal and photodegradation of soil organic matter from wheat field substrate. *Journal of Geophysical Research: Biogeosciences* **1, pp. 54-62**.

Whitford WG. **1981**. Exceptions to the AET model: deserts and clear-cut forest (litter, decomposition). *Ecology* **62, pp. 275–277**.

Wong FKY, Lau MCY, Lacap DC, Aitchison JC, Cowan DA, Pointing SB. **2010**. Endolithic microbial colonization of limestone in a high-altitude arid environment. *Microbial Ecology* **4, pp. 689-699**.

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