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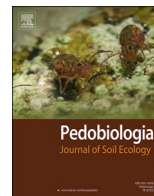
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## Taking sides? Aspect has limited influence on soil environment or litter decomposition in pan-European study of roadside verges

A. Amstutz<sup>a</sup>, LB. Firth<sup>a</sup>, JI. Spicer<sup>a</sup>, P. De Frenne<sup>b</sup>, L. Gómez-Aparicio<sup>c</sup>, BJ. Graae<sup>d</sup>, S. Kuś<sup>e</sup>, S. Lindmo<sup>d</sup>, A. Orczewska<sup>e</sup>, F. Rodríguez-Sánchez<sup>f</sup>, P. Vangansbeke<sup>b</sup>, T. Vanneste<sup>b</sup>, ME. Hanley<sup>a,\*</sup>

<sup>a</sup> School of Biological and Marine Sciences, University of Plymouth, Drake Circus, Plymouth, Devon PL4 8AA, UK

<sup>b</sup> Forest & Nature Lab, Department of Environment, Ghent University, Belgium

<sup>c</sup> Department of Biogeochemistry, Plant and Microbial Ecology, Institute of Natural Resources and Agrobiological of Seville (IRNAS, CSIC), Spain

<sup>d</sup> Department of Biology, Norwegian University of Science and Technology (NTNU), Trondheim, Norway

<sup>e</sup> Faculty of Natural Sciences, University of Silesia in Katowice, Poland

<sup>f</sup> Dept. Biología Vegetal y Ecología, Universidad de Sevilla, Spain

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### ABSTRACT

In addition to well-known effects on species ecophysiology, phenology, and distributions, climate change is widely predicted to impact essential ecosystem services such as decomposition and nutrient cycling. While temperature and soil moisture are thought to influence litter decomposition, elucidating consistent soil process responses to observed or predicted shifts in climate have proven difficult to evidence. Here we investigated how aspect (i.e., north-south orientation), a natural model for variation in soil temperature, influenced soil physico-chemical conditions and decomposition of two standardised litter types (Green tea and Rooibos teabags) in Pole-facing (PF) and Equator-facing (EF) roadside verges spanning a 3000 km and 27° latitudinal gradient across Europe. Despite average daily temperatures being 1.5 - 3.0 °C warmer on EF than PF slopes, there were only minor region-specific differences in initial soil physico-chemical conditions and short-term variation in litter decomposition (i.e., litter mass loss was higher in EF-verges for the first month of deployment only) associated with aspect. We conclude that previously observed differences in soil environments and the decomposition process associated with slope orientation, is largely litter or environment specific, although medium-term soil-decomposition in semi-natural grassland ecosystems may also be insensitive to the magnitude of temperature variation within the range predicted by the IPCC SSP1-2.6 emissions scenario. Nonetheless, consistent average and extreme temperature differences between adjacent PF- and EF-aspects along roadside verges provides a model system to explore exactly how resilient the soil environment and the micro-organisms responsible for decomposition, are to temperature variation.

### 1. Introduction

Decomposition is a fundamental biological process; it facilitates the recycling of chemical nutrients, and so helps maintain primary productivity, trophic interactions, and ultimately, drive the global carbon cycle (Wardle, 2002; Bradford et al., 2016). The breakdown and recycling of litter and detritus depends on various, often interacting, factors, including litter resource quality, soil biogeochemistry, the nature and activity of soil fauna and micro-organisms (Aerts, 2006; Walter et al., 2013; Bradford et al., 2016), but is widely held to be particularly

sensitive to variation in climate (Swift et al., 1979; Joly et al., 2023). Consequently, the environmental perturbations associated with Anthropogenic Climate Change (ACC) will likely have many direct and indirect effects on the decomposition process, and with it, important changes in soil organic carbon stores and nutrient cycling (Moinet et al., 2020; Parmesan et al., 2022).

Direct ACC-linked effects may manifest themselves through the influence of soil moisture and temperature on soil biology and chemistry (Hobbie, 1996; Aerts, 1997; Davidson and Janssens, 2006), but associated impacts on soil-organism abundance and activity are likely, with

\* Corresponding author.

E-mail address: [mehanley@plymouth.ac.uk](mailto:mehanley@plymouth.ac.uk) (ME. Hanley).

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multiple consequences for decomposition and nutrient turnover (Davidson and Janssens, 2006; Sierra et al., 2015). Nonetheless, observed effects of ACC-linked temperature changes on decomposition processes are extremely varied (Aerts, 2006; Allison and Treseder, 2011; Walter et al., 2013; Yin et al., 2019a; Homet et al., 2021). Although warming is predicted to facilitate a general upshift in decomposition through increases in soil enzyme, microbial and faunal activity (Aerts, 2006; Davidson and Janssens, 2006; Allison and Treseder, 2011), soil moisture has a strong, interactive relationship with warming, since reductions in soil moisture are usually associated with higher temperatures (Aerts, 2006; Walter, Sierra et al., 2013, 2015; Canessa et al., 2021). As a result, the activity of soil biota can decline sharply in warmer, drier soils (Allison and Treseder, 2008; Thakur et al., 2018; Yin et al., 2019b) and a concomitant reduction in decomposition rates ensues. This interaction, coupled with the confounding effects of land use variation, soil nutrient and organic matter status, litter quality, and seasonality (Aerts, 2006; Yin et al., 2019a; Joly et al., 2023), makes prediction of how these critical ecological processes will respond to likely ACC scenarios exceptionally difficult.

There is increasing realisation however, that climate-change experiments and predictive models need to embrace these complex, interactive effects if we are to understand ecosystem responses to ACC (De Frenne et al., 2013; Parmesan and Hanley, 2015; Parmesan et al., 2018; Yin et al., 2019a). Nevertheless, it becomes a logistical challenge to impose and control various ACC-linked factors in manipulative laboratory or field experiments (De Frenne et al., 2013; Parmesan and Hanley, 2015). One approach is to use so-called 'natural laboratories' to explore how species and ecosystems vary in response to climate factors. This includes studies conducted across large latitudinal gradients (De Frenne et al., 2013; Ma et al., 2019; Joly et al., 2023) and experiments using aspect as a local natural surrogate for often extreme climate variation (Nevo, 2012; Firth et al., 2016; Amstutz et al., 2021, 2024).

Latitude offers the advantage that variation in organismal traits can be set against a relatively predictable change in ambient mean annual temperatures (about 154 km °C<sup>-1</sup>), and to a lesser extent, average precipitation (De Frenne et al., 2013). For these reasons, many studies have evidenced important, if perhaps expected geographical patterns of community pattern and process from high to low latitudes (Pennings et al., 2009; Canessa et al., 2021; Jones et al., 2022; Joly et al., 2023). At a more local scale, habitat orientation to the sun ('aspect') can also provide significant variation in environmental conditions. Indeed, some studies (Mudrick et al., 1994; Sariyildiz et al., 2005; Jasińska et al., 2019) have reported a general trend for litter decomposition rates to be higher on Pole-Facing (PF) versus Equator-Facing (EF) slopes. While aspect no doubt has a tremendous influence on the identity and abundance of terrestrial organisms, and the roles they play in ecosystem processes (Badano et al., 2005; Warren, 2008; Cao et al., 2020; Patel et al., 2023), situations where natural processes have generated adjacent PF- and EF- slopes that can be used as controlled experimental pairs are limited. One of the best-known examples, is the 'Evolution Canyon' (EC) microscale model (Nevo, 2012), where despite the relative proximity of north- and south-facing aspects, very different ecosystems have developed on opposing slopes. This demonstrates how aspect-specific land-use and habitat differences, potentially developing over millennia, inevitably confound comparisons of ecological processes, even when adjacent sites share underlying geological and general climatic conditions.

Recent ecological studies on aspect conducted in the rocky intertidal have nonetheless revealed major biological differences between slopes. For EF- and PF-habitats separated by as little as two meters, species assemblages, trait expression, interactions, and within-species ecophysiology show major variation (Firth et al., 2016; Amstutz et al., 2021, 2024). Although intertidal rocky gullies are natural systems, such heavily disturbed habitats where propagule establishment is environment, rather than supply, restricted highlight the excellent potential that adjacent and recently disturbed sites have for elucidating the role of

aspect (and by inference, ACC effects) on biodiversity and ecosystem processes. We know of no comparable natural terrestrial equivalents, but roadside verges may offer broadly similar consistency and control of potentially confounding variables such as day-length, land-use, disturbance history, underlying geology, and the recruitment of species involved in key ecological processes at a scale of just a few meters.

Our aim was to determine how variation in environmental conditions on adjacent roadside verges influence decomposition on PF versus EF surfaces locally and combine this approach with an assessment of changes over a wide latitudinal range. We investigated how aspect affected litter decomposition and soil chemistry in four PF and EF road verge locations across each of five regions spanning a European latitudinal gradient of 27° (southern Spain to central Norway). In so doing, we investigated whether observed variation in decomposition rates on EF- versus PF- slopes (e.g., Mudrick et al., 1994; Sariyildiz and Küçük, 2008; Jasińska et al., 2019) were a habitat-specific or general phenomenon. In addition to quantifying temperature, soil moisture content, and the decomposition of standardised litter samples, we also considered variation in several other important soil characteristics (organic content, nitrogen and carbon content, pH) likely to affect the decomposition process (Aerts, 2006; Walter et al., 2013; Bradford et al., 2016; Jasińska et al., 2019). Specifically, we tested the following hypotheses:

- (1) Verge temperatures (mean, minimum, maximum) differ according to aspect but variation between EF and PF slopes remains consistent over broad geographical (Latitudinal and Longitudinal) scales.
- (2) Soil physico-chemical factors (i.e., soil moisture, pH, organic matter, Carbon and nitrogen content) differ according to aspect but any variation remains consistent over broad geographical (Latitudinal and Longitudinal) scales.
- (3) As a result of observed aspect-related variation in (1) and (2), the decomposition of standardised litter samples (teabags) will vary according to aspect, but any PF vs EF differences remain consistent over broad geographical (Latitudinal and Longitudinal) scales.

## 2. Methods

### 2.1. Study sites and plot location

We chose five different regions along a > 3000 km latitudinal gradient across Europe (see **SoI Table 1**); the most northerly was Trondheim (63°N) Norway, running progressively south to Arcos de la Frontera, Spain (36°N). As noted by de Frenne et al. (2013), the relationship between latitude and mean March-September temperatures is - 0.65 °C per degree of latitude in the Northern extratropical hemisphere. This would yield an expected temperature difference during the main growing season of 17.6 °C between Trondheim and Arcos, although in practice, local factors including topography, wind speed, radiation balance, cloud and snow cover and atmospheric moisture all affect local temperature and precipitation regimes. Additionally, the Belgian (Ghent), Polish (Katowice), and English (Plymouth) regions, were situated at similar latitudes (50°N), almost equidistant between Trondheim and Arcos de la Frontera and consequently, offered sampling points over a wide longitudinal range (i.e., 22° from Plymouth to Katowice).

At each of the five regions we selected four paired mesic grassland locations that offered long sections of opposite PF and EF roadside verge habitat sloping at an angle of approximately 45°. Within each verge location, three parallel 1 m × 4 m plot areas were haphazardly selected, but with the proviso that they were at least 1 m from the road edge or other disturbed, shaded or rocky area. Within each, we marked three 0.5 m × 0.5 m sub-plots, 1 m apart and positioned at the same relative height and distance from the road as sub-plots in the paired site opposite (see **SoI Fig. 1**). We assumed that the relative proximity of opposing,

paired roadside verge locations would limit any confounding effect of time since, or the nature of, road construction, or the impacts of traffic load and anthropogenic disturbance on soil conditions.

## 2.2. Variation in microclimate, soil organic content, and chemistry

Data loggers (HOBO UA-002-08 Pendant Temp/Light, Onset Computer Corporation, Bourne, MA, USA), attached to a vertical bamboo cane 5 cm above ground level, were deployed to record (hourly) air temperature for every PF and EF-slope location. Any vegetation within a 1 m radius of each logger growing above the height of the logger was cut back to prevent shading. We recorded temperatures for the 3-month duration of the decomposition trials at each site (Spring 2018).

At the start of the experiments, we collected 400 g of surface soil (i.e., from the 'A-horizon', ~3–15 cm deep) from an area immediately adjacent to the 1 m × 4 m plots. All stones, roots, and macro-organisms were removed, and material bagged for transport. Three sub-samples (~50 g) were used locally to determine soil moisture content for each slope. These samples were sieved (< 2 mm), weighed and oven-dried at 105 °C until they achieved constant mass, the mass difference used to quantify the gravimetric soil moisture. The remaining soil samples (250 g each) of sieved (< 2 mm) dried soil (50–60 °C overnight) for each slope location across the European transect were sent to Plymouth for analysis of pH, nitrogen, carbon and organic matter content. Soil pH was measured in sub-sample (90 g, for 3 replicates of 30 g, from each slope) of soil dissolved in an equal volume of deionized water using a glass electrode (YSI Pro 1030, Yellow Springs, Ohio, USA). Nitrogen and carbon were measured by coal pyrolysis analysis (i.e., helium chromatography followed by flash combustion in an oxygen rich environment - EMASyst., 1996). Organic matter content was quantified using the dry combustion method (4 hrs at 650 °C in a muffle furnace, see Goldin, 1987).

## 2.3. Litter Decomposition

We followed the standardised assay method of Keuskamp et al. (2013) using commercially available tea bags to assess variation in litter decomposition. Two different types of tea were used to provide different litter qualities, namely *Lipton Sencha Green Tea* which has low C:N ratio and is relatively labile, and *Lipton Rooibos* which has a high C:N ratio and is more recalcitrant (Keuskamp et al., 2013; Djukic et al., 2018). Teabags were deployed by removing (as one piece) the top 5 cm of plant material and soil in each of the 0.5 m × 0.5 m sub-plots within the three 1 m × 4 m plots, spacing four green tea and four rooibos tea bags at regular distances on the soil surface. Once done, the teabags were re-covered with the intact earth sod.

The timing of initial teabag deployment depended on region, and (for Norway) snow cover. Consequently, deployment times varied by around two months between Spain (early March) and Norway (early May). We recovered (four) tea bags from one of the (0.5 m × 0.5 m) sub-plots within each of the three (1 m × 4 m) plots at each site sequentially, one, two, and three months after deployment.

## 2.4. Statistical analyses

Annual daily mean, maximum and minimum values for each region were calculated and the effect of 'Aspect' (PF- or EF-) and 'Region' (Norway, Belgium, Poland, England & Spain) on each was examined using a two-way ANOVA using the slope averages from each location as replicate units (i.e.,  $N = 4$ ). Each of the soil abiotic parameters (carbon and nitrogen content; soil moisture; pH; organic matter content) quantified from the three replicate soil samples collected for each plot were pooled to calculate plot averages. Following logit transformation to account for non-normality in proportional data (Warton & Hui 2011), the effect of 'Aspect' (PF- or EF-) and 'Region' (Norway, Belgium, Poland, England & Spain) on each parameter was examined using a two-way ANOVA.

Teabag decomposition was estimated by quantifying the percentage mass loss of individual tea bags during field deployment, and then calculating the average mass loss for all 12 tea bags per slope location recovered after 1-, 2- and 3-months exposure. Following logit transformation, we used two-way ANOVA to compare the interactive effect of 'Region' and 'Aspect' on mean tea bag decomposition, using all four aspect locations in each region as replicates.

## 3. Results

### 3.1. Microclimate and soil environment

In addition to a highly significant 'Region' effect we found consistently higher mean daily temperatures on EF slopes across the European gradient, varying from an average 3 °C difference in Belgium, to 1.5 °C in Spain (Table 1). While we also detected similar trends for mean daily minimum and maximum temperatures, the magnitude of difference for these parameters was considerable. Only Norwegian sites (1.5 °C) showed an average minimum temperature difference for PF- vs EF sites more than 0.7 °C, while the minimum difference in average maxima (Spain) was never below 6.1°C, and for the Belgian sites, mean maxima were 13 °C higher on EF. In all cases, the absence of any significant 'Region' × 'Aspect' interaction highlighted that EF slopes are consistently warmer than PF slopes.

Although, all soil environmental factors varied greatly across the five regions, none showed any consistent variation with 'Aspect', nor was there any significant 'Region' × 'Aspect' interaction (Table 2). Verges in England consistently had the highest Organic matter, soil N, and moisture content, but the lowest observed soil pH. There was less apparent variation in soil conditions between the other four regions, but verges in Spain had the highest soil pH and carbon content.

**Table 1**

Variation in average daily mean, mean minimum and mean maximum ( $\pm$  SE), above-ground temperatures recorded on Pole- (PF) and Equator-facing (EF) roadside verges for five regions located across Europe (Norway, Belgium, England, Poland, and Spain). Results of two-way ANOVA (following logit transformation) comparing the individual and interactive effect of 'Aspect' and 'Region' on each thermal parameter are also shown.  $N = 4$  paired PF and EF verge slope locations for each region except Poland and Belgium where one data logger was lost on one PF-slope for each region.

Region	Mean Daily Temperature (C)		Mean Daily minimum Temperature (C)		Mean Daily maximum Temperature (C)	
	PF	EF	PF	EF	PF	EF
Norway	16.47 $\pm$ 0.20	19.19 $\pm$ 0.07	6.77 $\pm$ 0.08	8.22 $\pm$ 0.38	27.30 $\pm$ 0.56	38.85 $\pm$ 0.87
Belgium	16.08 $\pm$ 0.57	19.03 $\pm$ 0.09	9.57 $\pm$ 0.26	9.47 $\pm$ 0.16	23.91 $\pm$ 2.24	36.91 $\pm$ 0.55
England	13.00 $\pm$ 0.35	15.92 $\pm$ 0.55	5.52 $\pm$ 0.68	6.00 $\pm$ 0.73	23.05 $\pm$ 0.09	34.20 $\pm$ 1.20
Poland	17.48 $\pm$ 0.49	19.88 $\pm$ 0.64	7.46 $\pm$ 0.79	8.18 $\pm$ 0.18	30.68 $\pm$ 0.24	40.04 $\pm$ 1.03
Spain	18.50 $\pm$ 0.14	20.06 $\pm$ 0.17	8.96 $\pm$ 0.18	9.56 $\pm$ 0.07	32.70 $\pm$ 0.47	38.80 $\pm$ 0.03
<b>Two-way ANOVA<sub>(DF)</sub></b>	<b>F</b>	<b>P</b>	<b>F</b>	<b>P</b>	<b>F</b>	<b>P</b>
Aspect <sub>(1,28)</sub>	103.35	< 0.001	5.94	< 0.001	183.00	< 0.001
Region <sub>(4,28)</sub>	47.71	< 0.001	25.92	< 0.001	14.25	< 0.001
Interaction <sub>(4,28)</sub>	1.34	0.358	0.69	0.620	2.33	0.800

**Table 2**

Variation in mean ( $\pm$  SE), soil organic matter and moisture content, pH, carbon (C), and nitrogen (N) associated with Pole- (PF) and Equator-facing (EF) roadside verges located in five regions across Europe (Norway, Belgium, England, Poland, and Spain). Results of two-way ANOVA (following logit transformation) comparing the individual and interactive effect of 'Aspect' and 'Region' on each parameter are also shown.  $N = 4$  verge slope locations for each region.

Region	Organic matter (% dry mass)		pH		Moisture (%)		C (% dry mass)		N (% dry mass)	
	PF	EF	PF	EF	PF	EF	PF	EF	PF	EF
Norway	5.3 $\pm$ 0.5	5.2 $\pm$ 0.4	6.7 $\pm$ 0.2	7.3 $\pm$ 0.2	33.6 $\pm$ 1.9	26.1 $\pm$ 2.1	2.2 $\pm$ 0.9	2.3 $\pm$ 0.7	0.1 $\pm$ 0.02	0.1 $\pm$ 0.02
Belgium	5.4 $\pm$ 0.2	5.6 $\pm$ 0.2	6.9 $\pm$ 0.3	6.9 $\pm$ 0.2	37.1 $\pm$ 1.3	35.3 $\pm$ 1.4	1.7 $\pm$ 0.4	2.0 $\pm$ 0.5	0.1 $\pm$ 0.01	0.2 $\pm$ 0.01
England	12.5 $\pm$ 0.6	12.7 $\pm$ 0.7	5.4 $\pm$ 0.2	5.0 $\pm$ 0.1	55.9 $\pm$ 3.4	48.1 $\pm$ 3.7	4.9 $\pm$ 2.0	5.5 $\pm$ 2.0	0.4 $\pm$ 0.02	0.5 $\pm$ 0.03
Poland	5.9 $\pm$ 0.4	10.5 $\pm$ 1.2	8.2 $\pm$ 0.3	7.9 $\pm$ 0.3	25.5 $\pm$ 1.5	24.8 $\pm$ 1.4	3.9 $\pm$ 0.7	6.8 $\pm$ 1.1	0.2 $\pm$ 0.02	0.3 $\pm$ 0.05
Spain	7.6 $\pm$ 0.5	7.8 $\pm$ 0.8	9.3 $\pm$ 0.03	9.5 $\pm$ 0.04	28.6 $\pm$ 0.8	30.6 $\pm$ 1.1	7.0 $\pm$ 0.8	7.4 $\pm$ 0.8	0.1 $\pm$ 0.01	0.1 $\pm$ 0.01
<b>Two-way ANOVA<sub>(DF)</sub></b>	<b>F</b>	<b>P</b>	<b>F</b>	<b>P</b>	<b>F</b>	<b>P</b>	<b>F</b>	<b>P</b>	<b>F</b>	<b>P</b>
Aspect <sub>(1,30)</sub>	1.53	0.225	0.03	0.867	2.14	0.154	2.76	0.107	2.09	0.159
Region <sub>(4,30)</sub>	13.38	< 0.001	31.97	< 0.001	33.11	< 0.001	23.28	< 0.001	32.26	< 0.001
Interaction <sub>(4,30)</sub>	1.36	0.271	0.70	0.596	0.54	0.708	0.77	0.551	1.76	0.164

### 3.2. Aspect and Tea bag decomposition

For brevity, we do not show the results or analysis of tea bag mass loss after 2-months, as patterns were identical to those described for the 3-month deployment (see [SoI Table 2](#)). When tea bags were recovered after 1-month field exposure ([Fig. 1](#)), we not only found significant variation in mass loss across the five regions (One-month 'Region' effect Rooibos  $F_{4,30} = 11.81$ ,  $P < 0.001$ ; 'Region' Sencha  $F_{4,30} = 7.74$ ,  $P < 0.001$ ), but a trend for consistently higher mass loss on EF-slopes (One-month 'Aspect' effect Rooibos  $F_{1,30} = 5.10$ ,  $P = 0.031$ ; 'Aspect' Sencha  $F_{1,30} = 4.85$ ,  $P = 0.035$ ). There was no significant 'Region'  $\times$  'Aspect' interaction (One-month Rooibos  $F_{4,30} = 1.20$ ,  $P = 0.33$ ; Sencha  $F_{4,30} = 0.011$ ,  $P = 0.59$ ), suggesting a broadly consistent 'Aspect' effect across latitude (although Sencha mass loss on PF-slopes in Norway was slightly higher than on EF- (i.e., 39.9 vs. 39.6% respectively - [Fig. 1](#))).

While the 'Region' effect remained after 3-long month exposure (Three-month 'Region' effect Rooibos  $F_{4,30} = 11.85$ ,  $P < 0.001$ ; 'Region' Sencha  $F_{4,30} = 12.55$ ,  $P < 0.001$ ), we failed to detect any consistent difference in tea bag mass loss with aspect (Three-month 'Aspect' effect Rooibos  $F_{1,30} = 0.89$ ,  $P = 0.115$ ; 'Aspect' Sencha  $F_{1,30} = 0.52$ ,

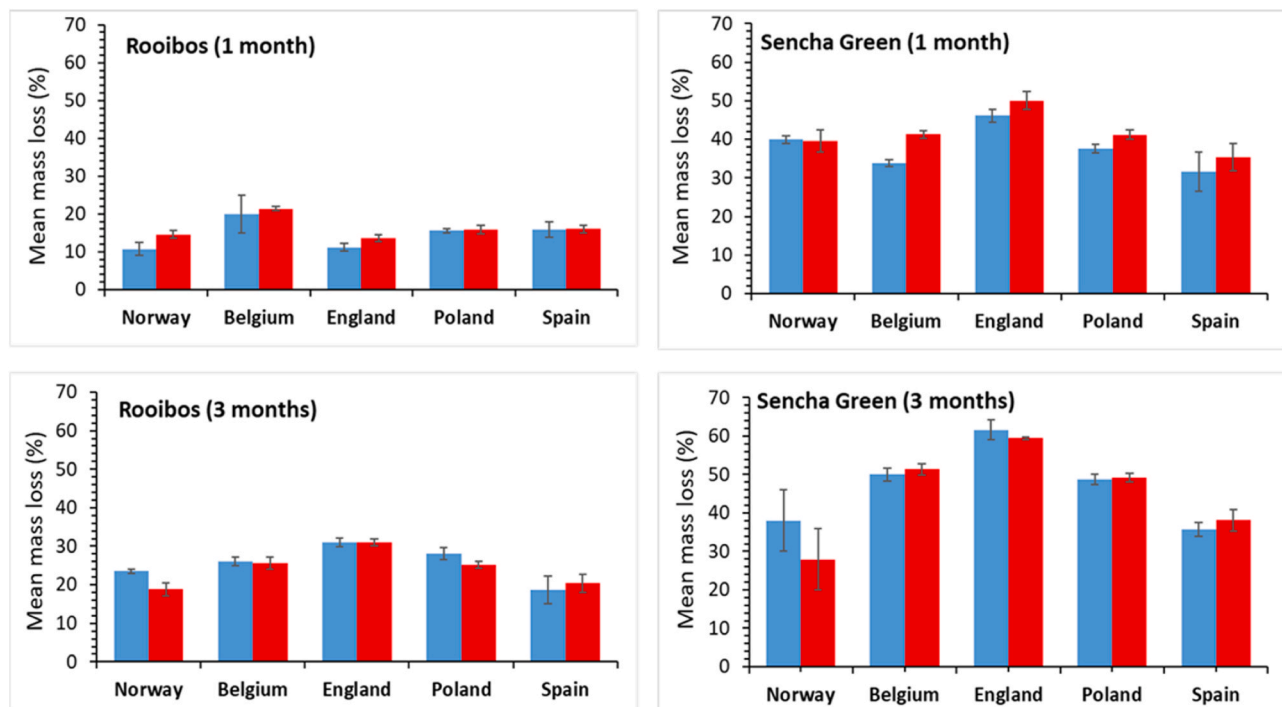
$P = 0.474$ ), or a significant 'Region'  $\times$  'Aspect' interaction (Three-month Rooibos  $F_{4,30} = 1.29$ ,  $P = 0.296$ ; Sencha  $F_{4,30} = 1.18$ ,  $P = 0.339$ ).

Generally, Sencha Green Tea consistently had around two-fold greater mass loss than Rooibos throughout the experiment, and for 1-month deployment, the most marked differences in mass loss between aspects (about 4% more in EF-slopes - Norway excepted - [Fig. 1](#)). Especially for Sencha Green Tea, mass loss at the locations in England was greater than the other four regions, while tea bags in Spain tended to have the lowest mass loss.

## 4. Discussion

### 4.1. Overview

Our study is the first large-scale, multi-region (in total, twenty EF-versus PF-slope comparisons performed across Europe) investigation of the influence of slope aspect on soil physio-chemical factors and litter decomposition. The variation in average daily temperatures for PF-versus EF slopes correspond well to the 1.5 °C - 2.0 °C global temperature rise (above pre-industrial baseline) predicted for the end of this



**Fig. 1.** Variation in mean ( $\pm$  SE) percentage mass loss of Sencha Green Tea and Rooibos tea bags deployed for one- or three-months in Pole- (Blue) and Equator-facing (Red) roadside verge locations situated in five regions across Europe (Norway, Belgium, England, Poland, and Spain).  $N = 4$  paired verge slope locations for each region.



century under the IPCC SSP1–2.6 emission scenario (Lee et al., 2021). Also relevant to our understanding of how ecological processes like decomposition might respond to the higher temperature associated with ACC (Parmesan and Hanley, 2015; Parmesan et al., 2022), we observed marked aspect-related variation in mean maximum temperatures (i.e., usually over 10 °C). Nonetheless, despite these consistent, Europe-wide differences in thermal environment between PF- and EF-slopes, and an expectation that such temperature differences would influence decomposition and therefore, nutrient cycling (Aerts, 2006; Allison and Treseder, 2011; Parmesan et al., 2022), there was limited support that the soil environment or decomposition process varied accordingly.

#### 4.2. Effect of aspect on decomposition

Our discovery of consistently higher teabag mass loss on EF-slopes after 1-month deployment contrasts with Mundrick et al., (1994), working in the US Appalachian Mountains, Sariyildiz et al., (2005) in Northeast Turkey, and Jasińska et al., (2019) working in an inland dune system in northern Poland. All three studies reported greater litter decomposition in PF-slopes. Interestingly however, when Jasińska et al., (2019) contrasted litter losses in early versus later successional dune plots, they also reported greater reductions in EF-slopes. Our sites were relatively well-established grassland habitats, so in this case, an early successional influence seems unlikely. Jasińska et al., (2019) ascribed their site-specific differences to variation in microbial decomposer activity and the initial chemical properties of the local litter samples used, but like Mundrick et al., (1994) and Sariyildiz et al., (2005) did not establish a clear causal link between decomposition and local environmental conditions, except to suggest that soil moisture was consistently greater on PF-slopes.

Nonetheless, it seems reasonable to speculate that soil moisture might influence litter quality and benefit the microbial and arthropod decomposer communities involved in decomposition (see Suseela et al., 2012; Sierra et al., 2015). We detected no aspect-linked variation in soil moisture. Nonetheless, the much greater mass loss of Sencha tea bags observed in England, where soil moisture (amongst other factors) was also much elevated compared to other regions, highlights the importance of this factor generally. It is possible that a combination of relatively high soil moisture and warmer conditions on EF-slopes early in the respective field seasons for each region was sufficient to stimulate the soil microbial community and facilitate enhanced decomposition for a short period until the soils started to dry out. Potentially, PF-slopes conserved higher soil moisture longer in the growing season resulting in a higher decomposition that compensated the initial lag compared to EF-slopes.

Our inability to detect any ‘Aspect’ effect at 3-month deployment is nonetheless, in marked contrast to Mundrick et al., (1994), Sariyildiz et al., (2005) and Jasińska et al., (2019). All three studies reported greater litter decomposition on PF-slopes in experiments lasting from 1 to 3 years, and in the case of Mundrick et al., (1994), for samples taken every 2-months throughout their year-long experiment. This difference may simply reflect the fact that all our 2- and 3-month deployment samples were recovered between May and August when soil moisture levels had perhaps decreased to a point where microbial activity on both aspects was below a critical level for decomposition (Aerts, 2006; Walter et al., 2103, Sierra et al., 2015; Canessa et al., 2021).

#### 4.3. Importance of litter type

Another major difference between our study and the earlier experiments by Mundrick et al., (1994), Sariyildiz et al., (2005) and Jasińska et al., (2019) is that rather than deploy locally sourced leaf litters, we used a standard tea bag litter assay (Keuskamp et al., 2013; Djukic et al., 2018). It is possible that the nature of the litter material is important in detecting environmental influence on decomposition. Like most large-scale experiments of this kind (e.g., Berg et al., 1993; Joly et al.,

2017; Djukic et al., 2018), we used a standard litter type (teabags) rather than locally sourced materials. A recent pan-European study by Joly et al. (2023) however, suggests that the influence of major macroclimatic variation on decomposition is moderated by litter quality and type. Specifically, when decomposition of litter ‘native’ to a particular region was compared to standard litter, macroclimatic variation exerted a significant influence on decomposition of the former, but not the latter. Joly et al. (2023) concluded that the use of standard litters like teabags might lead researchers to over-estimate the influence of local microsite variation over any site-specific climate effect, since decomposition was likely greatly influenced by the local decomposer community and their preference for, and ability to deal with, native and non-native litter types (so-called ‘home-advantage’; see also Franzitta et al., 2015). The more labile and Nitrogen-rich Sencha Green Tea decomposed much faster than the Carbon-rich, recalcitrant Rooibos tea. While neither tea blend is likely to reflect closely the characteristics of naturally occurring ‘native’ litter in any of our regions, this difference highlights how litter quality, and especially Nitrogen content, influences the rate of decomposition, if not emergent patterns between different aspects or regions.

While our one-month-long teabag deployment did support a consistent ‘macroclimate’ effect, the results of our longer-term tea bag deployments corroborate Joly et al.’s (2023) assertion that major climate-linked patterns can only be elucidated using native litters. This may also be why Mundrick et al., (1994), Sariyildiz et al., (2005) and Jasińska et al., (2019) all found consistently greater decomposition in PF-slopes over several years.

#### 4.4. Implications for the decomposition process in a climate change world

Notwithstanding our initial (1-month) observation, our 2- and 3-month sample results may signal an optimistic conclusion that despite consistent differences in average mean daily and mean maxima air temperatures between PF- and EF-aspects, soil-based patterns and processes in semi-natural grassland ecosystems are relatively well-buffered from the climate shifts expected with the lower range of IPCC (Lee et al., 2021) emissions scenarios. This may seem counter-intuitive, but a recent study by Robinson et al. (2022) conducted in Iceland shows that microbial decomposers were unexpectedly resistant to soil warming, even over a temperature gradient spanning 10–35 °C. We recognise however, that a complex suite of interacting factors can obscure predicted impacts of ACC on ecological relationships. More generally, there is widespread acceptance that ACC has influenced greatly the biology of many different species (Parmesan et al., 2022) and consequently, species distributions and resulting ecological interactions such as decomposition and nutrient cycling will inevitably be disrupted (Kirwan and Blum, 2011; Steidinger et al., 2019). Nonetheless, it is evident that we currently understand much less about the complexities of these critical community-level, ecological processes than we do species-specific responses, even though we cannot expect to comprehend the biological impacts of ACC without it (Parmesan and Hanley, 2015; Parmesan et al., 2018, 2022).

While we found only short-term differences in tea bag mass loss, by providing proximal, ‘paired’ locations where confounding variables such as day-length, land-use and disturbance history, underlying geology, and propagule supply are relatively controlled, roadside verges (across latitudinal gradients) may nonetheless, offer a window into how decomposition responds to climate variation (Swift et al., 1979; Joly et al., 2023). Future studies should, however, use locally sourced litter materials (Joly et al., 2023) deployed for longer periods (and to detect seasonal variation), and consider aspect-linked variation in soil micro-organism communities and activity to better elucidate and explain any emergent decomposition patterns.

#### Declaration of Competing Interest

The authors declare that they have no known competing financial

interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data Availability

Data will be made available on request.

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## Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at [doi:10.1016/j.pedobi.2023.150927](https://doi.org/10.1016/j.pedobi.2023.150927).

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