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USING THE TEA BAG INDEX TO MEASURE DECOMPOSITION RATES IN ICELANDIC RANGELANDS

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ABSTRACT

This study was conducted in Audkuluheidi and Theistareykir where grazing is practiced during summer. The tea bag index (TBI) method was used to measure decomposition rates in those Icelandic highlands with exclosure experiments with the following objectives: (1) to estimate decomposition rates in grazed and non-grazed areas, in heath and gravelly desert in Audkuluheidi and Theistareykir, and (2) to learn the method of TBI for measuring decomposition rates. Initial weights for tea bags were taken before installation, then tea bags were taken to the field and buried in pairs (green and rooibos) at a depth of 5 cm in the experimental plots, organised in six pairs in two habitats. That part of the work was done by other members of the research team. The tea bags were collected in June 2022 after 12 months of incubation and dried in an oven for 48 hours at 70°C. The tea bags' final weights were recorded and analysed to determine the effects of grazing exclusion, habitats, and sites on mass loss of the two tea types. The results showed that there were significant differences between habitats and sites in tea mass loss. There was no significant difference in mass loss in grazed/non-grazed plots, meaning that grazing did not have any effect on decomposition processes. By using TBI it is possible to collect information about land health through measuring decomposition rates in order to identify strategies for sustainable management of ecosystems, such as rangelands.

Key words: decomposition rate, degradation, Icelandic highlands, tea bag index, rangelands

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1. INTRODUCTION

Decomposition of organic matter plays a crucial role in the proper functioning of ecosystem processes such as cycling of energy for heterotrophic organisms, nutrient cycling, and regulation of hydrology (Gupta & Singh 1977). The process of decomposition is the means through which minerals held in the tissue of plant structures are transferred to the soil for reuse by plants (Haynes 1986). The process of decomposition represents a link in the nutrient cycle of ecosystems (Swift et al. 1979). Decomposition is controlled by internal factors, such as litter quality and microorganism community structure, as well as external factors, like moisture, temperature, and soil pH (Swift et al 1979; Preston et al. 2012). Decomposition can be altered by disturbances caused to the ecosystem, such as grazing, fires, over cultivation and landslides, among others. Due to decomposition, carbon is released back into the atmosphere as carbon dioxide while nitrogen (in mineralised form) is made available in a form that can be easily absorbed by plants (Haynes 1986).

One of the main sources of organic matter that serves as substrate for decomposition in terrestrial ecosystems is plant litter (Krishna & Mohan 2017). When litter decomposes it becomes a major source of fixed carbon in terrestrial ecosystems and has the ability to maintain its structure and function over time in the face of external stress (Ashworth et al. 2021). The litter above and below-ground in terrestrial ecosystems constitutes the main energy resource for a variety of soil organisms through complex interactions (Hättenschwiler et al. 2005). The breakdown of litter in an ecosystem requires both physical and chemical processes to reduce organic matter material to its elemental chemical components (Aerts 2006). During organic matter turn over, the main organisms that are involved in biomass breakdown are soil bacteria and fungi (Hättenschwiler et al. 2005). The process of litter decomposition could be summarized in two steps (Aerts 2006). In the first step, plant material is reduced by the help of detritivores to tiny fragments which can be chemically reduced (Aerts 2006). In the second step, bacteria and fungi chemically convert those tiny fragments into ammonium, phosphate, water, and carbon dioxide (Swift et al. 1979).

Being such an important process in ecosystem functioning, decomposition could be used as an indicator of ecosystem function (Tiegs et al. 2013). To date, litter bags containing native plant litter are still being used to measure decomposition rates and drivers for decomposition. Most studies normally use litter locally collected within the same environment for measuring decomposition rates. Using locally available plant litter can be good in delivering realistic results for decomposition for a particular area, but it comes with some challenges (Björnsdóttir 2018). For example, litter collected locally decomposes more quickly within that environment compared to litter brought from other environments (Ayres et al. 2009). Such effects can bring bias and can interfere with comparisons between studies and sites (Didion et al. 2016). Keuskamp et al. (2013) introduced a new method for measuring decomposition using tea bags, the tea bag index (TBI). This method is simple and can be used for comparisons of decomposition rates in different ecosystems and soils (Björnsdóttir 2018). Simplified and commercially available Lipton tea bags, which contain dead plant material, are used to measure decomposition rates. Two types of tea are used, green and rooibos. They have varying quantities of labile and recalcitrant fractions and thus varying decomposability. The two tea types are installed as one pair (Björnsdóttir 2018).

Many ecosystems, such as rangelands, are facing degradation because of increasing numbers of grazing livestock (Pulido et al. 2017), which is the case for the Icelandic highlands. Problems associated with degradation processes due to large numbers of livestock and heavy grazing are decline in vegetation cover and compaction of soils (Pulido et al. 2018). Overgrazing can cause excessive vegetation loss leading to a reduction in the overall productivity of rangelands (Boone et al. 2018). When there is a substantial decrease in vegetation cover due to overgrazing, disruption of ecosystem processes such as the hydrological cycle, nutrient cycling and energy flow can lead to reduced resilience and integrity of the ecosystem (Mayor et al. 2013).

In this project, the tea bag index method was used to measure decomposition rates in different stages of degradation in Icelandic rangelands where grazing occurs during summer. Use of grazing exclosure is a potential way to restore rangelands and can effectively reduce degradation and enhance vegetation growth in overgrazed areas (Wang et al. 2018), but if the level of degradation has passed some ecological threshold, exclosure alone cannot be enough for the revegetation of such rangelands (Sasaki et al. 2013).

In Uganda, land degradation is mainly due to inappropriate agricultural methods such as deforestation, bush burning and overgrazing (Pender et al. 2004). The high rate at which land is degraded in Uganda is due to poverty, high population growth and land fragmentation leading to over exploitation of land. Bagoora (1988) indicated that the highest rate of degradation is mostly observed in the highlands. Finding ways to turn around these trends is very important for Uganda. For that to be successful, information about land health, for example through measuring decomposition rates, is needed to help identify strategies that will lead to more productive and sustainable land use. The TBI method can be used to collect data to inform management practices for most ecosystems.

1.1 Objectives and research questions

The overall goal of this study was to use the TBI method to measure decomposition rates of land in different conditions in the Icelandic rangelands and to learn the method in order to apply it in Uganda.

Specifically, the objectives were:

1. To estimate decomposition rates in grazed and non-grazed areas, in relatively intact and degraded ecosystems (heath and gravelly desert) and between two sites (Audkuluheidi and Theistareykir) using the TBI.

2. To learn the TBI method of measuring decomposition rates in order to adapt it to different contexts and studies in Uganda.

The research questions were:

1. Do decomposition rates vary between grazed and non-grazed sites in the Icelandic highlands?

I expected to find higher decomposition rates in non-grazed plots compared to grazed plots because without grazing the aboveground biomass is expected to increase, whereas with grazing the aboveground biomass is disturbed and may lead to changes in the soil system (Mulloy et al. 2021).

2. Do decomposition rates vary between relatively intact and degraded ecosystems in Icelandic highlands?

Here, I expected to find higher mass loss in heath than in melur (the Icelandic term for a gravelly desert) because the soil surface in melur is exposed and there is therefore a higher chance for erosion that may affect decomposition (Hättenschwiler et al. 2005).

3. Do decomposition rates vary between sites located outside (Audkuluheidi) or inside (Theistareykir) the volcanic active zone in Iceland?

I expected to find a higher decomposition rate in Theistareykir than Audkuluheidi because the decomposition rate tends to be higher in areas with young volcanic soils (Berenstecher et al. 2017).

2. METHODS

2.1 Study area

The study was conducted in two sites in the Icelandic highlands (Fig. 1A): Audkuluheidi in the central highlands, 470 m above sea level (65.13 N 19.67 W) and outside the volcanic active zone, and Theistareykir in the north-east, 380 m above sea level (65.89 N, 17.08 W), within the volcanic active zone (Mulloy et al. 2019). The dominant type of vegetation at these highland sites is subarctic-alpine tundra vegetation that experiences very cold winters and short summer seasons for vegetation growth. Environmental conditions at the two sites are broadly similar, but there are also some differences. Theistareykir lies at lower elevation and since it is closer to the coast, it has a more oceanic climate, while Audkuluheidi is more continental.

Even though the two highland areas experience almost the same climatic conditions, the weather information captured by the weather stations of the Icelandic Meteorological Office (IMO 2022) around the two sites for the period between 2012 and 2022 indicate that there are some differences in climate (Table 1).

Table 1. Weather information for Audkuluheidi and Theistareykir showing the average in climatic conditions for the years 2012 to 2022. (Source: IMO 2022).

	Audkuluheidi	**Theistareykir**
Annual mean temperature	0.92˚C	2.01˚C
Average summer temperature from June-August	7.98 ˚C	8.65˚C
Average relative humidity	83.9%	79.7%

Updated precipitation data for the two highlands was not found but data from 1994-2003 showed annual precipitation to be 707 mm and 657 mm for Audkuluheidi and Theistareykir respectively (IMO 2022).

Audkuluheidi lies on a basaltic bedrock and has mainly well-drained brown Andosols and glacial deposits uphill (Arnalds 2015). In Theistareykir, the soils are well aerated basaltic Andosols. Andosols have a carbon content of about 2% and clay containing allophane (about 22.5%) and ferrihydrite (about 4%). These soils have high cation exchange capacity and water holding capacity of up to 45% (Arnalds 2004). Soil pH at the study sites is neutral (around 5-6) and their carbon to nitrogen ratio is usually low, therefore favouring plant growth (Jónsdóttir et al. 2005; Akello 2019).

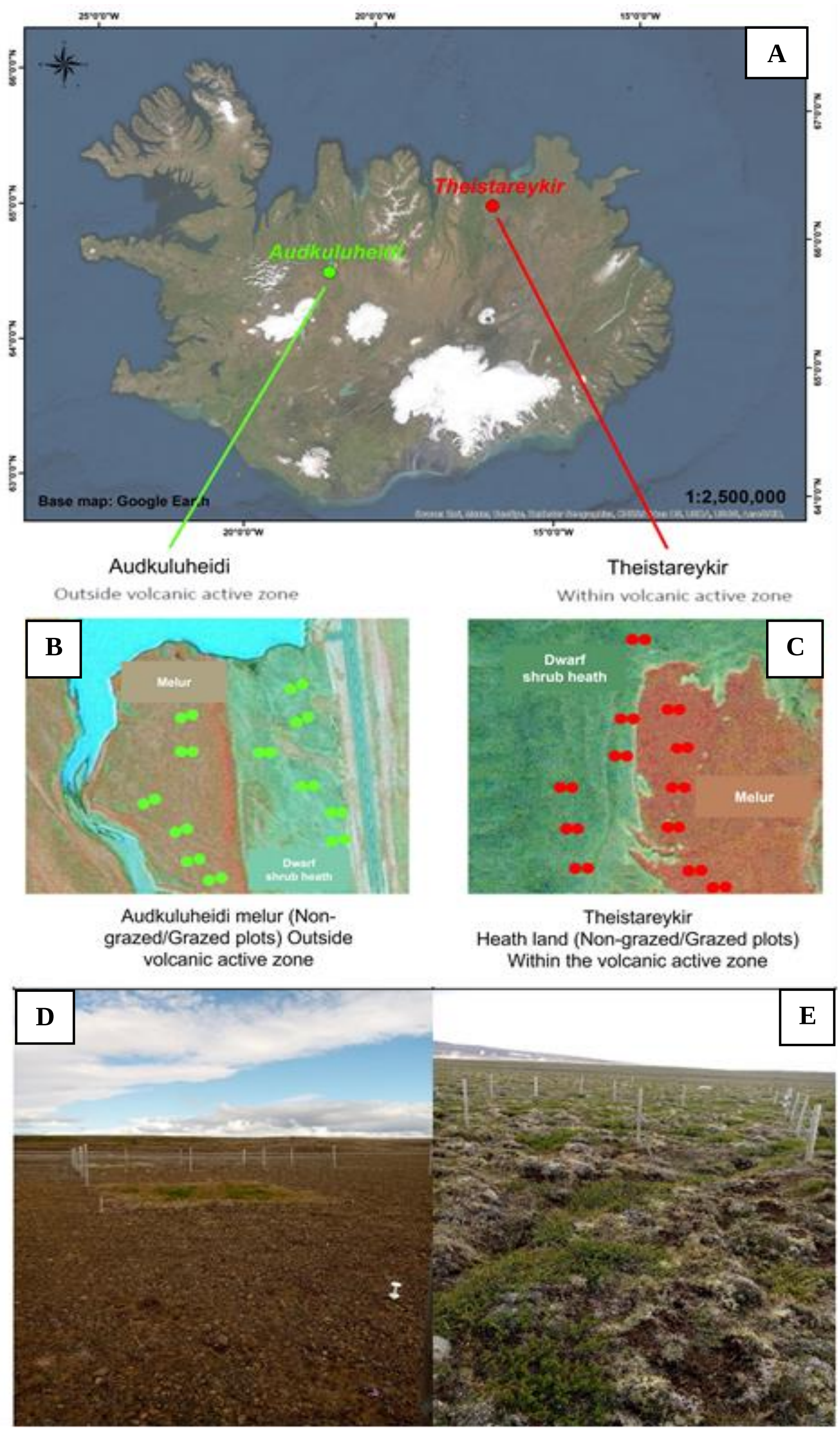

Figure 1. (A) Audkuluheidi situated outside the volcanic active zone and Theistareykir within the volcanic active zone. At each site (B & C), six pairs of grazed and non-grazed areas were established in two habitats (melur and heathland. D & E show photos of degraded (D) and vegetated (E) habitats and plots.

Landscapes at these sites are characterized by a mosaic of habitat patches in different stages of degradation. This includes well-vegetated heath of more than 70-90% plant cover with dwarf shrub, dominated by *Betula nana*, and a high abundance of bryophytes and lichens (Jónsdóttir et al. 2005), as well as sparsely vegetated areas locally referred to as melur with less than 5-10% vegetation cover of forbs and graminoids and highly degraded by grazing and wind erosion (Sierro Miguel 2017). Extensive sheep grazing is practiced during summer in both study sites (Thórhallsdóttir 1997).

The melur and heathland habitats are assumed to have been similar at one time but, because of erosion by wind and degradation by grazing, they changed to what they look like today (Mulloy et al 2021). The two habitats represent two extremes of a degradation gradient, from the heathland representing a healthier ecosystem to the gravelly desert representing a collapsed ecosystem (Mulloy et al 2021).

2.2 Experimental design

A field experiment was set up at the two sites in 2016. Experimental plots of 12 m x 12 m in size were set up in heath and melur habitats (24 plots in each habitat). Plots were organized in six pairs for each habitat/site as shown in Fig. 2. Within each experimental pair, the fencing treatment was allocated at random, 100 m apart from the next pair and 4 m between them. The fencing posts were made of metal and wood, 1 m in height with a spacing of 4 m and enclosed with a net wire of 20 cm x 10 cm to prevent sheep from grazing within the enclosures. The non-fenced plots were left intentionally open to allow free grazing by sheep during the summer.

The measurements of decomposition rates presented in this study were taken in 2022 with tea bags that were installed in 2021 within the field experiment described above.

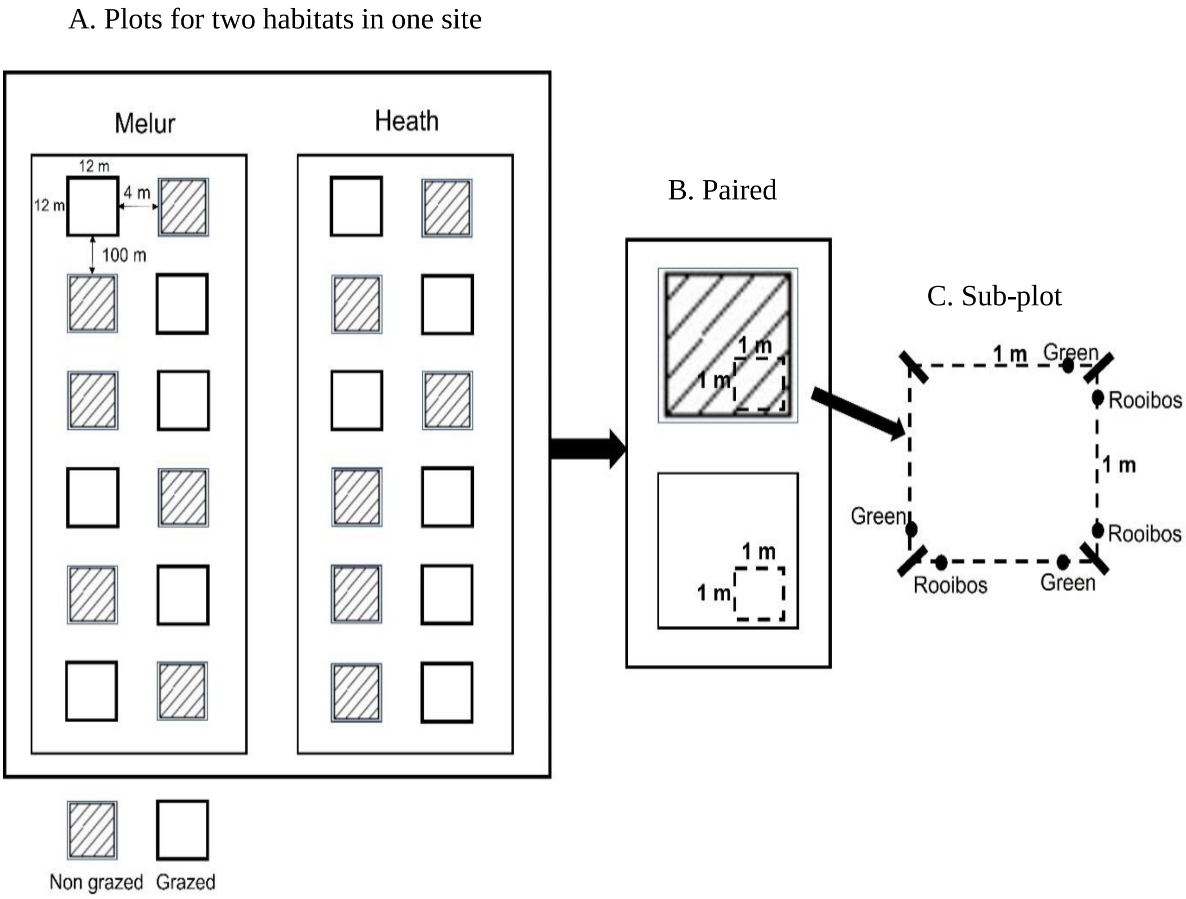

Figure 2. Experimental design for the study indicating (A) six pairs of grazed and non-grazed plots at each study site in two habitats of heath and melur (only one site represented), giving a total of 24 plots for each site, (B) one pair of grazed and non-grazed plots of 12 m x 12 m and (C) sub-plot of 1 m x 1 m with black dots at the three corners where the tea bags were installed in pairs.

2.3 The TBI method

2.3.1 Material in the tea bags

To measure decomposition rates, commercially available tetrahedron-shaped synthetic tea bags containing appr. 2 g of green tea or rooibos tea (Fig. 3A) were used. The green tea has a high content of labile carbon, and the rooibos tea a high content of recalcitrant carbon. The mesh of 0.25 mm that encloses the tea allows microorganisms and mesofauna to enter the bags but excludes macrofauna (Setälä et al. 1996).

2.3.2 Preparations and installation

Before taking the tea bags to the field, their initial weights were taken and each tea bag was labelled with the plot and corner identifier on the back of the tea bag label. The tea bags were taken to the field and buried in pairs (green and rooibos) at a depth of 5 cm in three corners of each 1 m x 1 m sub-plot in the experimental grazed and non-grazed plots. Each pair was marked with a small wooden pin so that it could be easily found during collection. This part of the work was done by other members of the research team.

2.3.3 Tea bag collection

The tea bags were collected on 16th and 17th June 2022 from the two described study sites after 12 months of incubation. The bags were carefully collected using spoons, as shown in Fig. 3B. Each pair of tea bags (from each corner) was placed in a separate Ziploc bag, together with a small piece of paper including information on the plot identity and corner from which the pair of tea bags was retrieved. This allowed identification of the tea bags even when the label was no longer readable or was lost. All the Ziploc bags from one plot were put into a separate plastic bag. The tea bags were then transported to the soil laboratory at the Agricultural University of Iceland and kept refrigerated (5°C) until further processing.

Figure 3. Tea bags and the process of retrieving them from the field. (A) **C**ommercially available synthetic tea bags used for TBI measurements. (Photo: Keuskamp et al. 2013) (B) tea bags being carefully collected using a spoon to prevent breaking the bags and destroying the soil system where tea bags were installed.

2.3.4 Mass loss estimation

In the laboratory, the tea bags were gently brushed to remove adhered soil particles from the outer surfaces (Fig. 4A). Roots growing in the tea bags were cut just outside the bag, making sure not to force them out by pulling. The tea bags with their paper tags were gently packed together on small aluminium boxes to avoid mixing up tea bags from different plots (Fig. 4B).

The tea bags were placed to dry in an oven for 48 hours at 70°C (Fig. 4C) and their weights recorded. The aluminium boxes with tea bags were randomly picked one at a time from the oven, to ensure they would not absorb any moisture that could affect the readings of the weights of the tea bags. Finally, the weights of string, label, and empty bags of 5 green tea and 5 rooibos tea bags were randomly picked after general weights were taken to calculate the exact mass (litter) lost from green tea and rooibos tea bags (Fig. 4D).

Figure 4. Showing the processes in the laboratory. (A) gentle cleaning of adhered soil particles and cutting of the roots that had entered the tea bags during incubation in the field, (B) tea bags were packed in pairs with a paper tag denoting their plot in small aluminium boxes, (C) boxes containing the tea bags were placed in an oven for 48 hours at 70°C, and (D) the tea bags were weighed and their weights recorded.

2.4 Statistical analysis

To analyse potential differences in mass loss as a function of grazing, location (Audkuluheidi and Theistareykir), or habitat type (well vegetated heath and sparsely vegetated melur), I conducted multiple one-way analyses of variances (ANOVAs). All statistical analyses were conducted using R, version 4.2.1 (R Core Team, 2022).

3. RESULTS

3.1 Decomposition of different types of organic matter at grazed and non-grazed sites

Mass loss of green tea in grazed plots (mean ± SE: 1.01 ± 0.02 g) was almost two times higher than that of rooibos tea (red tea) in grazed plots (mean ± SE: 0.53 ± 0.02 g). The mass loss of green tea in non-grazed plots (mean ± SE: 1.03 ± 0.02 g) was also higher than that of rooibos tea in non-grazed plots (mean ± SE: 0.55 ± 0.01 g). However, after 12 months of incubation in the field, there was no significant difference in mass loss between grazed and non-grazed plots, either for green tea (F=0.90, df=1,14, p=0.35; Fig. 5) or rooibos tea (F=1.17, df=1,14, p=0.28 Fig. 5).

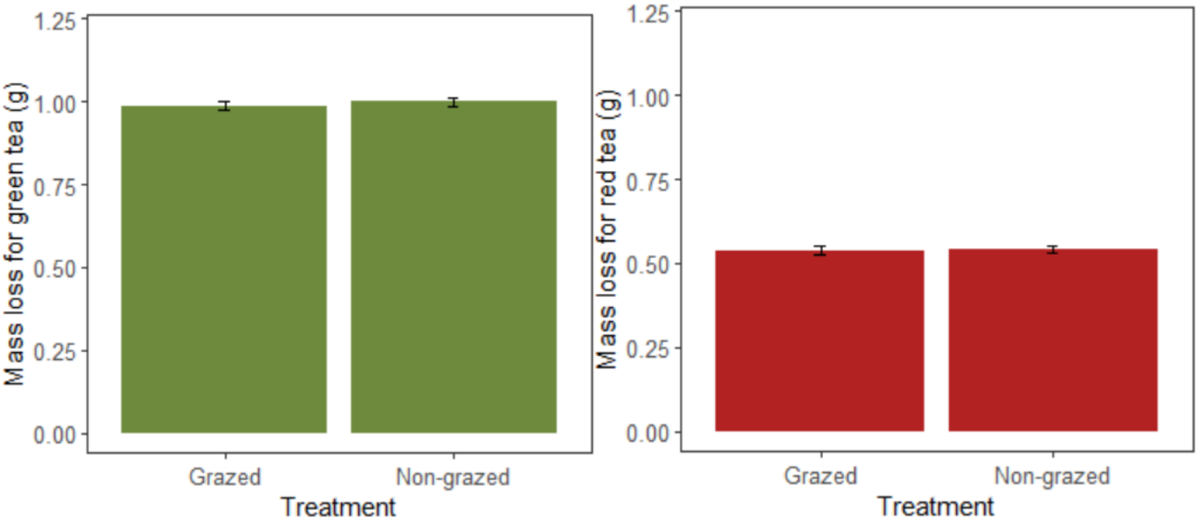

Figure 5: Mass loss of green tea (left) and rooibos (red) tea (right) in grazed and non-grazed experimental plots. Bar heights represent mean values of mass loss in different treatments using green and rooibos tea bags, together with the standard error.

3.2 Comparison between habitat types

In heath vegetation, the mass loss of green tea (mean ± SE: 0.96 ± 0.01 g) was almost two times higher than that of rooibos tea (mean ± SE: 0.56 ± 0.02 g). For melur, mass loss of green tea (mean ± SE: 1.09 ± 0.02 g) was two times higher than that of rooibos tea (mean ± SE: 0.52 ± 0.01 g). There was significant difference in mass loss between heath and melur with green tea (F=45.77, df=1,14, p<0.001 Fig. 6) and with rooibos tea (F=5.10, df=1,14, p=0.03 Fig. 6).

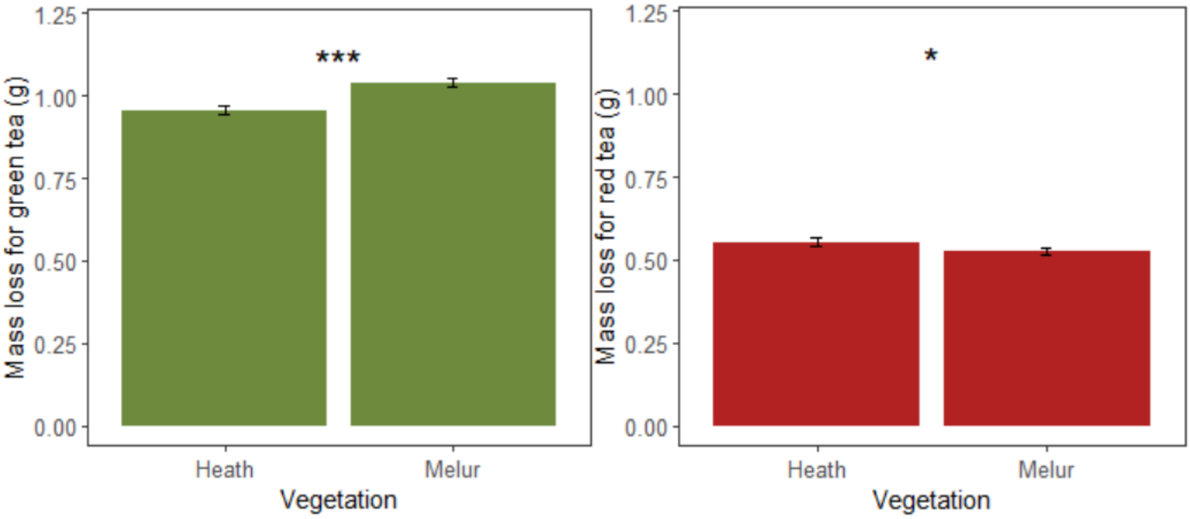

Figure 6: Summary of the effects of habitat type (heath and melur) on decomposition using green tea (left) and rooibos (red) tea (right). Asterisks indicate significant differences between the mass loss in the two habitats (* p<0.05; *** p<0.001).

3.3 Comparison of decomposition rates between Audkuluheidi and Theistareykir sites

In Audkuluheidi, the mass loss of green tea (mean ± SE: 0.98 ± 0.01 g) was almost two times higher than rooibos tea (mean ± SE: 0.52 ± 0.02 g), while in Theistareykir, mass loss of green tea (mean ± SE: 1.07 ± 0.02 g), was two times higher than for rooibos tea (mean ± SE: 0.56 ± 0.01 g). After 12 months of incubation of the tea bags, there was a strong significant difference in mass loss between Audkuluheidi and Theistareykir for green tea ($F=20.66$, $df=1,14$, $p<0.001$ Fig. 7), but not for rooibos tea, which showed no difference in mass loss between habitats ($F=3.11$, $df=1,14$, $p=0.08$ Fig. 7).

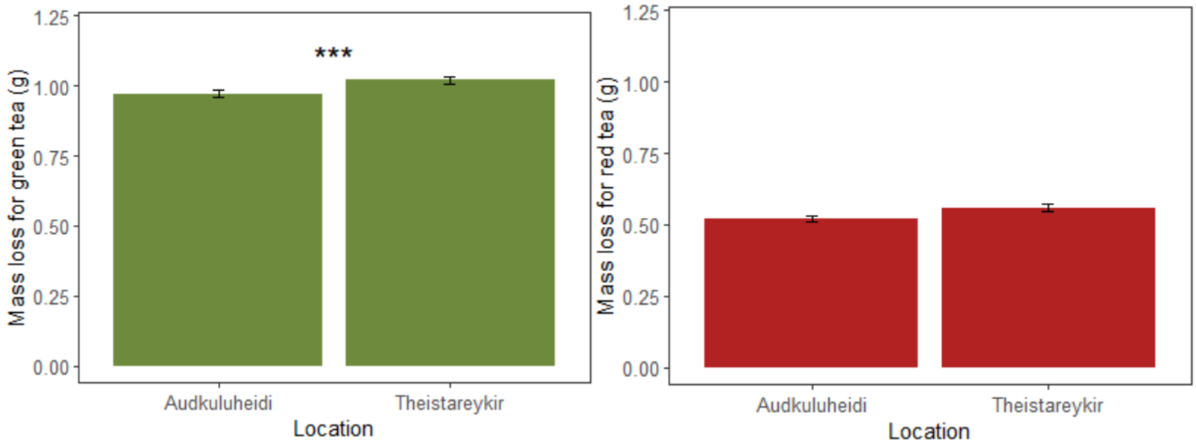

Figure 7: Comparison of decomposition rates between sites: Audkuluheidi and Theistareykir for green tea (left) and rooibos (red) tea (right). Asterisks show significant differences between the site within the volcanic active zone and the site outside the volcanic active zone using green tea (*** p<0.001).

4. DISCUSSION

The results of this study showed that the TBI can be used to assess decomposition, which is a main ecosystem process, under different conditions. Specifically, this method was used to assess the effect of different treatments (grazing/non-grazing), vegetation (heath/melur) and locations (within the volcanic active zone/and outside of the volcanic active zone) on mass loss of tea types with varying quantities of labile and recalcitrant fractions. The results showed that there were significant differences between vegetation (heath/melur) and location (Audkuluheidi/Theistareykir) in tea mass loss after 12 months incubation of the two tea types. However, there was no significant difference in mass loss between grazed and non-grazed plots.

The lack of differences between grazed and non-grazed plots was contrary to my expectation of mass loss in non-grazed sites being higher than in grazed sites, based on the assumption that grazing alters the composition and function of rangeland structure of both the aboveground biomass and soil structure (Bardgett & Wardle 2003; Mulloy et al. 2021). However, the results indicate that grazing did not have any effect on mass loss of either green tea or rooibos tea. This could be because grazing usually occurs for only a few summer months and that could enable quick recovery of aboveground biomass after the summer, hence may not significantly affect mass loss (Sun et al. 2016). Several studies have shown that litter type affects the decomposition rates because different litter types attract different microbial communities causing differences in decomposition (Aerts 1997; Hättenschwiler & Jørgensen 2010; Didion et al. 2016; Dossou-Yovo et al. 2021).

The results showed higher mass loss for green tea in melur than in heath, but the opposite pattern was found for rooibos tea. The expectation was to find higher mass loss in heath for green tea because in melur the soil surface is exposed, leading to higher chances of erosion that may affect decomposition (Hättenschwiler et al. 2005). The mass loss for rooibos was higher in heath than in melur which could be because soils in vegetated plots harbour certain groups of microorganisms, especially fungi, which are responsible for complex processes of decomposition (Lu et al. 2017). The differences between heath and melur using green tea could have been due to soluble compounds in green tea that could have leached out more in melur due to the sandy nature of the soils that allows quick water circulation, thus leading to higher mass loss in melur than in heathland (Jensen et al. 2005). Rubino et al. (2010) noted that observed differences in decomposition rates could be linked to changes in microbial community and compounds held in the litter (Lu et al. 2017).

Mass loss was higher in Theistareykir than in Audkuluheidi (Fig. 7), although it was expected that mass loss would be lower in the volcanic active zone due to the periodic deposition of ash on the soils. This contrary result could be because certain types of organisms are not affected by volcanic ash (Berenstecher et al. 2017). The low mass loss in Audkuluheidi probably implies unfavourable conditions, such as low temperatures and high moisture content, that could have affected microbial activity because highly moist soils affect oxygen supply to the microorganism thereby affecting aerobiosis (Schinner 1982). Rooibos tea showed no significant differences in mass loss. This could be because labile carbon-loving microbes were affected differently by proximity to the volcanic active zone than recalcitrant carbon loving microorganisms.

5. CONCLUSIONS

This study showed that a grazing exclusion of six years did not significantly affect the decomposition processes of litter in the highlands of Iceland. Still, grazing alters vegetation structure and composition, and these effects need to be considered when developing management strategies for rangeland ecosystems. Due to the short duration of the grazing season in Iceland (3 months), the effects caused by herbivory could have been undetected in this study since microbial activity happens slowly in colder areas. More research should be conducted to determine the average time frame needed for grazing to have effects on decomposition processes in the Icelandic highland rangelands.

In Uganda, degradation is mainly due to conversion of wetlands, grasslands, rangelands, shrublands and forests to crop fields, thus affecting the integrity of the ecosystems. Finding ways to turn around these trends is important and information about decomposition rates is needed to help identify strategies that will lead to more productive and at the same time sustainable land use. The intention of this study was to learn the TBI method and to be able to apply it back in Uganda since it is simple, the tea bags are commercially available and results can be easily comparable across the globe.

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