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Decomposition in differing snow regimes in high Arctic Svalbard

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DOI for data sets and R scripts

All data sets used in the analysis for this thesis, and the corresponding R script, can be found by following this link:

https://doi.org/10.18710/WRUJVF

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The above document contains:

- Readme txt file with all meta information
- Dry mass loss of tea bags at collection dates in 19 June 2018 and 27 August 2018 for annual, winter, summer, and summer OTC data sets
- Soil surface temperature outside OTCs from September 2017 to August 2018, logging hourly, as downloaded from Tinytag loggers, calibrated for zero shoulder offsets, and to a 0°C curtain
- Overview of calibration offset numbers for Tinytag loggers outside OTCs, using a 0°C water bath in September 2017
- Soil surface temperature inside OTCS from June 2018 to August 2018, logging every half our, as downloaded from Tinytag loggers, unaltered and not calibrated
- Soil moisture measurements from 19 June 2018 and 1 September 2018
- R-scripts for decomposition, temperature, and soil moisture analysis
- Plot map of fences with data loggers (updated August 2018)
- Schematic diagram with overview of tea bag and OTC placement behind snow fences

Foreword

Before you lies my master thesis, "Decomposition in differing snow regimes in high Arctic Svalbard", written between September 2018 and May 2019, for the BIO-3950 Master's Thesis of Biology in Northern Populations and Ecosystems, at UiT the Arctic University of Norway.

The research questions were formulated in collaboration with my supervisors, Prof. Elisabeth Cooper and Dr. Martin Mörsdorf. Dr. Mörsdorf began the initial phases of this experiment in September 2017, before I began my thesis study in June 2018.

I would like to thank my supervisors for their guidance and expertise during this process. Special thanks to Prof. Cooper for funding my fieldwork, and to Dr. Mörsdorf for his patience and compassion while I struggled to adjust to the dark winter months in the Arctic. I also wish to thank Eva Breitschopf (UiT), Mikel Moriana Armendariz (UiT), Tomoki Morozumi (Hokkaido University), and Trine-Lise Slåtsveen (UiT) for fieldwork assistance and for their excellent attitude despite the rain. Thank you to the University Centre in Svalbard for allowing me to use their lab facilities, and to the safety team for providing daily weather and polar bear updates. Thank you to UiT for the use of their lab facilities, for my office space, for funding, and for the opportunity and privilege of studying in Norway.

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1 Abstract

Rapidly rising temperatures are having great effects on the high arctic ecosystem, historically characterized by long, cold winter periods, and cold dry summers. Due to the length of the winter season, arctic soils have been an important carbon sink, but may now be facing increased microbial activity due to increasing temperatures. Temperature, moisture, and litter quality play important roles in decomposition processes, where winter temperature, summer moisture, and plant community composition is strongly influenced by the amount of accumulated snow during the winter period. Increased winter snow depth has been found to increase winter soil temperatures, therefore increasing microbial activity and decomposition in the winter period. Decomposition of green and rooibos tea bags was studied in a long-term experiment in Adventdalen, Svalbard, where snow was manipulated behind snow fences to create Ambient, Medium, and Deep regimes. Open top chambers (OTCs) were used to measure simulated warmer summer temperatures on tea bag decomposition. Decomposition was measured for an annual term (September 2017-August 2018), a winter term (September 2017-June 2018), and a summer term (June 2018-August 2018, inside and outside of open top chambers). Temperature loggers recorded soil surface temperature hourly throughout the study period. Enhanced snow depth increased winter temperatures in the *Deep* and *Medium* snow regimes compared to Ambient. The Deep regime had delayed snowmelt compared to the Ambient and Medium regimes, and was coolest regime during the summertime. The Medium regime was the warmest regime during summer, by visual observation. Soil moisture decreased in all snow regimes over the course of the summer period, and soil moisture measured inside of OTCs was lower than outside of OTCs. Green tea and rooibos tea demonstrated different patterns of decomposition under the varying snow depth regimes. Annual decomposition was lowest in the *Deep* regime for both substrates. Annual decomposition was highest in *Medium* for green tea and highest in *Ambient* for rooibos tea. Decomposition was lower inside of OTCs than outside of OTCs for both types of tea. These results demonstrate the complexity of the processes driving decomposition in Arctic soils. Increased snow cover may cause lower decomposition in long-term studies where plant and microbial community composition may have changed over time, suggesting that winter soil temperatures alone may not be as important in these changing ecosystems.

2 Introduction

2.1 Future Arctic climate scenarios

The arctic ecosystem is very sensitive to climate change, and is experiencing rapid and severe changes with anthropogenic-induced rising global temperatures (ACIA, 2004). Historically, due to long winter seasons with low soil temperatures, the arctic tundra has been an important carbon sink, characterized by low decomposition rates (Björkman et al., 2010; Gorham, 1991; Oechel et al., 1993). Furthermore, due to the length of the winter season, winter soil respiration in the arctic makes up a significant portion of the annual carbon cycling (Mikan et al., 2002; Schimel et al., 2004). Ongoing climate changes in the Arctic could have an effect on various dynamics of this malleable system. A possible feedback loop has been predicted as a future climatic dynamic wherein increased snow cover results in increased soil temperatures during the winter, leading to more nutrient cycling via increased winter microbial activity. Shifts in precipitation patterns may be observed due to higher moisture levels in the atmosphere with increasing temperatures, leading to changes in snow cover during the wintertime, and an earlier onset of snowmelt in the springtime (Post et al., 2009). The amount of snowfall has been increasing in the Eurasian Arctic over the past century and has been predicted to continue to rise in some areas, while other areas, like the North American Arctic and arctic maritime regions, may see a decrease in snowfall (Bintanja et al., 2017; Callaghan et al., 2011). Changes in snow cover, both increasing and decreasing, affect soil temperature, soil moisture, and consequently nutrient cycling in these ecosystems. Deeper winter snow cover has shown increased carbon dioxide efflux during the wintertime, paired with decreased net carbon dioxide efflux during the summertime (Welker et al., 2000). These observed and predicted changes in arctic snow regimes will therefore have significant and dynamic consequences on decomposition and tundra ecology.

2.2 Decomposition processes and environmental conditions

Decomposition is the process by which dead organic matter is broken down by way of leaching, fragmentation, and chemical reduction into carbon dioxide (CO₂) and other inorganic compounds (Chapin et al., 2002). Soluble materials from the decomposing organic matter is leached by percolating liquids, such as rainfall and snowmelt, while large soil invertebrates fragment materials into usable forms for soil microbes and smaller soil fauna. Dead organic matter is chemically altered primarily by bacteria and fungi through biotic processes, and is impacted by soil moisture and snow cover insulation (Gavazov, 2010). The microbial decomposition process starts when water-soluble

compounds and non-lignified carbohydrates are decomposed first due to their more biologically available nutrients. This creates a relatively rapid initial rate of decomposition at the introduction of dead organic matter, but the processes begin to slow down once the decomposition of lignin begins; the remaining litter fractions decompose concurrently at a more constant rate (Berg, 2000). The last phase of decomposition occurs when the lignin fraction stabilizes, and the remaining dead organic matter becomes more humified and recalcitrant, and relatively unavailable to soil microbes (Berg, 2000).

Cumulatively, these processes create a homogenized substrate of soil organic matter (SOM), which constitutes a large part of the arctic ecosystems carbon and nitrogen pools (Gavazov, 2010). This SOM is largely composed of recalcitrant material, where the majority of the dead organic material has been broken down and cycled through the nutrient cycling system, available for plant uptake. Turnover of SOM in permafrost soils is driven by a complex relationship between temperature, moisture, oxygen, nutrient availability, and soil forming factors (Walz et al., 2017). Due to low temperatures and poor soil drainage, water logged soils in the Arctic create anaerobic conditions, which further inhibits decomposition processes, and much of the organic soil layer in arctic soils consequently still contains a largely labile portion of only partially decomposed organic matter (Walz et al., 2017; Weintraub et al., 2003). Therefore, changes in ambient summer air conditions and snow depth in the winter could have a great effect on microbial activity due to changes in temperature and moisture of arctic soils. Increased winter snow depth has been found to increase microbial activity (Morgner et al., 2010), specifically under certain vegetation regimes (Semenchuk et al., 2015), and depending on the decomposers in question (Mundra et al., 2016). Furthermore, most plant communities are nitrogen limited in the tundra, but the amounts of biologically available carbon and nitrogen still dictate the microbial demand for nitrogen, and the consequent rate of nitrogen mineralization. Changes in plant community composition, by way of climate change, would change the composition of the detritus pools in these soils. The litter type and quality has a great effect on microbial mineralization, and changes in the plant community can therefore have an important impact on microbial activity (Weintraub et al., 2003).

2.3 Increased snow fall and consequent changes in soil temperature and moisture

It has been found that increases in snow depth, resulting in increases in winter soil temperature, produces changes in ecosystem respiration and nutrient cycling (Morgner et al., 2010; Mörsdorf et al., 2019; Semenchuk et al., 2016, 2015). The responses of plant litter decomposition to climate

change are critical in understanding future ecological conditions in the Arctic. Microbial decomposition of plant litter is an important step in the cycling of nutrients within the soil system. Changes in winter snow regimes—combined with higher summer temperatures— affect the onset of snowmelt in the springtime, and soil moisture and soil temperature during the summer period (Cooper et al., 2011).

Since soil microbial activity is sensitive to changes in temperature and moisture, variation in future snow depth regimes and snow melt timing could have great importance. However, the relative importance of temperature and moisture, and the nature of these relationships with decomposition, is still uncertain (Sierra et al., 2015). Snow fences constructed in Adventdalen, Svalbard, were established in 2006 to simulate an ecosystem with increased accumulation of snow cover. These fences were used for this study to investigate the potential effects of environmentally changed snow regimes on decomposition. For this we used green and rooibos tea bags, as a standardized proxy for plant litter, following the methodological protocol established by the global Tea Bag Index (Keuskamp et al., 2013).

2.4 The Tea Bag Index

The Tea Bag Index (TBI) (<u>http://www.teatime4science.org/</u>) is a multifunctional and wellstandardized method of measuring decomposition that can be applied globally, in different climates and soils, where tea bags are buried in the soil following a strict protocol. The TBI uses two tea plant materials; rooibos tea and green tea, which consider the complex chemical composition of different plant litter and the decay of cellulose and other plant constituents that affect the interactions between litter and microbes. As soil microbial activity is also sensitive to the quality and type of litter, the use of rooibos and green tea represents differences in substrate qualities between plant species (Cornelissen et al., 2007). Although this method cannot replace the precision of local plant litter methods, it is cost effective, requiring few resources by using commercially available tea bags, and has been tested for sensitivity and robustness in various contrasting ecosystems and soils (Keuskamp et al., 2013). Green and rooibos tea bags are used in this experiment to identify variations in decomposition, using a measurement of mass loss, that are affected by varying winter snow depth and warmer summer conditions.

The purpose of this study is to quantify annual and seasonal decomposition of tea plant material in the high Arctic under various potential future environmental scenarios. The design considers temperature, soil moisture content, snow cover, and litter substrate. I aim to assess the following questions: 1) how do varying depths of snow cover affect winter and summer decomposition of rooibos and green tea plant material in the Arctic; 2) how do enhanced summer temperatures affect the decomposition of these tea bags and; 3) do enhanced summer temperatures interact with enhanced snow regimes to affect the decomposition during summertime?

2.5 Hypotheses

- I hypothesize that both types of tea plant litter will decompose most rapidly in the *Deep* (D) snow regime, slightly less rapidly in *Medium* (M), and most slowly in *Ambient* (A). I hypothesize this will be the case in both winter and summer, due to the insulation effects of snow cover during the wintertime, and increased soil moisture in the summertime from the snow cover during the winter period.
- I hypothesize that summer decomposition rates will be enhanced by increased summer temperatures inside open top chambers, due to warmer summer soil temperatures and increased microbial activity.
- 3) I hypothesize that enhanced summer temperatures will interact with enhanced snow regimes, where the predicted increased soil moisture in *Deep* combined with the increased summer temperature will result in the highest decomposition.

3 Methodology

3.1 Site Description

This study was conducted in Adventdalen (78°10'N, 16°06'E), a deglaciated river valley 12km East of Longyearbyen in central Spitsbergen, Svalbard. The Adventdalen flood plain drains westward into Adventfjorden, and is characterized by fluvial sediments, horizontally layered loess deposits, ice wedges, and continuous permafrost, which influences the water drainage in the area (Humlum et al., 2003). Peaks, ridges, and plateaus carved out by previous glacial events border this valley on the North and South sides, and further off towards the West. Because of the influence of warm North Atlantic currents on Spitsbergen, Adventdalen is milder than other inland Arctic regions found at the same latitude.

The study area occurs in the middle Arctic tundra zone, one of the five Arctic bioclimatic zones (based on July mean temperatures), where *Cassiope tetragona* (D. Don) heath and *Dryas octopetala* (L.)—*Tomentypnum nitens* (Hedw.) mesic meadow can be found (Elvebakk, 2005). The heath habitat usually procures on neutral to acidic substrates, with soils characterized by an increased

presence of stones. The heath is dominated by *Cassiope tetragona, Dryas octo-petala, Salix polaris* (Whalenb.), *Saxifraga oppositifolia* (L.), *Alopecurus borealis* (Trin.) and *Bistorta vivipara* (L.) (Cooper et al., 2011). In contrast, the meadow habitat is usually found on finer textured mesic calcareous substrates, and is dominated by *Salix polaris, Luzula arcuata* subsp. *confusa* (Lindeb.), *Alopecurus magellanicus* (Lam.), *Dryas octopetala* and *Bistorta vivipara* (Cooper et al., 2011). Morgner et al., 2010).

A common soil profile from this area, as described by Strebel et al. (2010), has an upper Ohorizon from 0.2 to 6 cm with an abundance of small roots and slightly decomposed organic plant matter. Following the O-horizon is a dark brown A-horizon from 1-5cm that sits above a B/C horizon of grey silt. Soil *in situ* pH values range from 5.0 to 6.5 moving down through the soil profile, characterizing a slightly acidic soil (Strebel et al., 2010). The soil in the heath plots of this study area have been recorded as shallow, rocky, and dry with a soil solution pH of 6.1, while the meadow blocks have a soil solution pH of 5.7 (Semenchuk et al., 2015). The 2347 year-old underlying permafrost soil in Spitsbergen is slightly acidic, with a recorded pH of 5.01, and has been in a frozen state for more than 1000 years (Hansen et al., 2007).

The closest meteorological station is located at Longyearbyen Airport, about 15km West of the study site. The annual mean temperature is -4.6°C (1981-2010 mean at Longyearbyen Airport; www.eklima.no). Annual total precipitation over this time period was 199mm, although due to strong winds and the lack of biomass on which the snow can be trapped to accumulate, it is likely that these presented values are low. Most precipitation is in the form of snow (www.eklima.no).

3.2 Methodology and Experimental Design

In the autumn of 2006, 12 snow fences were erected in Adventdalen as part of the SnoEco project (Morgner et al., 2010) covering an area of approximately 3.75km² to account for spatial and topographic variation. The fences are organized into four blocks (each block covering an area of approximately 200m x 200m, and at least 500m away from the next block), where each block has 3 fences (see Figure 1). Fences (1.5m tall, 6.2m wide) were built perpendicular to the predominant south-easterly winds, allowing snow accumulation on the leeward side of the fence (see Figure 2) (Cooper et al., 2011). As one fence was destroyed, eleven remaining snow fences were used for this study. Behind each snow fence, three snow depth regimes mark three treatment plots for this experiment: *Ambient* (A), *Medium* (M), and *Deep* (D) snow (Fig. 2, 3). The *Ambient* regime represents normal, unmanipulated snow cover (10-35cm deep) characteristic of most of the study area. *Deep* is directly behind the leeward side of the snow fence, approximately 3m from the fence, Page 6 of 47

and represents experimentally increased snow cover (approximately 150cm deep) with delayed onset of snowmelt. *Medium* snow depth represents an increase of snow cover to approximately 100cm deep, and is located approximately 10m from the fence. Snowmelt dates vary from year to year, but *Ambient* melts out before *Medium*, which melts out before *Deep* (Semenchuk et al., 2013 for more details). Snow depth was not measured in this experiment, but *Deep*, *Medium*, and *Ambient* snow depth regimes were pre-established in May of 2008, measured manually with an avalanche probe in 16 different positions behind the fence (Cooper et al., 2011; Semenchuk et al., 2016).



Figure 1. Aerial view of the Adventdalen snow fences, organized into blocks (A, B, C, D) with three fences per block (scale = approximately 3.75km²).



Figure 2. Depiction of wind direction in winter, and the consequent accumulation of snow on the leeward side of the snow fence. Snow depth corresponds to *Deep* (D) and *Medium* (M). *Ambient* snow regimes are not illustrated in this 2D figure, as they were always placed away from any snow fence manipulation.



Figure 3. Illustration for the experimental design and the location of tea bags buried in each snow regime and OTC treatment for all 11 snow fences in Adventdalen. The *Ambient, Medium,* and *Deep* snow regimes were not always placed in the same spot at each fence, but have been placed to insure correct location for the respective snow depth manipulation.

Experimental set up and data collection of this study was carried out in the summer months (September 2017, June 2018 and late August 2018) after spring snow melt and before the following winter's snowfall. A simplified litter experiment was carried out using commercially available Lipton green tea (EAN 87 10908 90359 5) and rooibos tea bags (EAN 87 22700 18843 8), following the methodological procedure from the Tea Bag Index (Keuskamp et al., 2013). The tea bags used in this experiment are tetrahedron in shape (5cm² sides), non-woven, and made from polypropylene material. Although the mass of the tea varies slightly for each tea bag, approximately 2g of tea is within each bag (89% green tea, 93% rooibos tea, both with supplemental natural flavouring). Green tea has a faster decomposition rate, and rooibos (red) tea has a slower decomposition rate, making it possible to concurrently assess decomposition (Keuskamp et al., 2013).

In spring 2017, tea bags were prepared to be buried in Adventdalen the following September. One tea bag from each box was weighed before and after being dried for 48hrs at 60°C to give a calibration mass for water loss from initial tea mass to after drying mass. The empty tea bag was also weighed and subtracted from the mass after drying to determine the initial mass of the tea. Each tag attached to the teabags was labelled with a unique ID number for identification. On September 06, 2017, following the stepwise TBI protocol (Teatime 4 Science), 3 rooibos and 3 green tea bags were buried in each snow regime (A, M, D) at each snow fence in Adventdalen to compare decomposition between snow depth regimes. Tea bags were buried 8cm deep into the soil, 15cm apart from one another, and 20cm East from Gemini Data Loggers (Tinytag, UK) that logged soil temperature every hour for the duration of the experiment (see Figure 4). Logger sensors were buried 1cm under soil surface in September 2017, but were later found sitting on the soil surface in August 2018.



Figure 4. Detailed burying scheme of tea bags in proximity to Tinytag temperature logger inside each snow depth treatment.

On 19 June 2018, one green tea bag and one rooibos tea bag was collected from each snow regime to give data for one winter season of decomposition (286 days of decomposition). Three volumetric soil moisture content measurements were taken beside each temperature logger in each snow regime (June soil moisture), using a handheld HH150 moisture meter and a SM150 sensor probe (Delta-T Devices Ltd, 2013). One new green tea bag and one new rooibos tea bag was buried in the place of the removed tea bags to give data for one summer season of decomposition. Open top chambers (OTCs) were set up beside each snow regime to give a corresponding OTC plot for snow regimes in order to synthesize a warmer summer climate with increased snowfall. New Tinytag loggers were planted on the soil surface inside the OTCs, set to record every 30 minutes for the summer period, and soil moisture was measured (June OTC soil moisture). Two new green and two new rooibos tea bags were also buried in the soil beside the loggers inside the OTCs, following the scheme in Figure 4, to give summer OTC decomposition data.

All Tinytag loggers, sensors, and cables were removed in 28 August 2018 and taken to Longyearbyen to be downloaded. One green and one rooibos tea bag was removed from each of the snow regimes on 27 August 2018 (annual decomposition data, 355 days) and the tea bags buried in June in each snow regime were removed (summer non-OTC decomposition data, 69 days). One green and one rooibos tea bag was removed from inside the OTCs (summer OTC decomposition data, 66 days). Soil moisture was measured inside each OTC (September OTC soil moisture) and outside OTCs (September soil moisture) on 1 September. OTCs were disassembled, temperature loggers, sensors, and cables were set back in the field as found (with sensors on the soil surface), and the remaining tea bags will make it possible to gather more data in the future.

Once removed from the field, sediment and fine roots adhering to the tea bags were removed, and the tea bags were dried in the laboratory of the University Centre in Svalbard for 24hrs at 35°C to stop decomposition, as suggested in the TBI protocol if drying at 60°C is not immediately possible. They were then transferred into individual bags and refrigerated until drying and weighing was possible in the Biology Lab at UiT The Arctic University of Norway. Once transported to UiT, tea bags were dried at 60°C for 48hrs, with any remaining soil and root debris removed from the outside of each bag, and cooled to room temperature (20°C) within the oven. Individual tea bags were weighed after drying (0.001g), and then cut open to remove the tea. Tea contents were weighed (0.001g) and recorded. Damaged tea bags were not used for analysis.

3.3 Data Analysis

3.3.1 Temperature

Gemini Data Tiny tag temperature loggers recorded temperature in degrees Celcius from 06 September 2017 to 28 August 2018. Logger data was extracted and transferred to excel files, where it was then processed in Rstudio. Non-OTC loggers (48 loggers) were calibrated using an ice bath in 2017, and checked for abnormal performance at this time, and individual loggers were checked for unusual performance in August 2018. OTC loggers (29 loggers) were not recently calibrated before use, so raw, unadjusted data was used for OTC temperature analysis. Temperatures were aggregated to evaluate mean daily temperatures, minimum daily temperatures, maximum daily temperatures, and mean daily temperatures below zero between the three snow regimes. Annual trends were evaluated by graphing logger data using the ggplot2 package, as well as trends during the melt out period, and during summertime. Temperature loggers inside OTCs were kept separate from non-OTC loggers for these descriptive analyses. Hourly temperature fluctuations from one day of data (day of year=200=19 July) for both OTC and non-OTC loggers was observed to remark differences in buffering effects of OTCs. Non-OTC loggers were calibrated after temperature extraction on past calibration information and to coincide with a 0° curtain.

3.3.2 Soil moisture content

Volumetric soil moisture content measurements were averaged (3 measurements for each snow depth regime (A, M, D) at each fence). Linear modelling was used to evaluate differences in soil moisture between treatments and between seasons. A likelihood ratio test and an Analysis of Variance (ANOVA), in combination with AIC values, were used to test the significance of an interaction between the effect of OTCs and snow regime treatments, resulting in the use of an additive model (June soil moisture ~ OTC + Snow regime) for June soil moisture and a multiplicative model for September (September soil moisture ~ OTC * Snow regime). Normality was visually determined by using a box plot to give an initial overview of the data, and a Q-Q plot of model residuals. Residuals were also visually checked for homoscedasticity. Outliers were kept or removed based on their "mildness" or "extremeness", as an attempt to determine their ecological importance using the quartile method described in Benhadi-Marín (2018). Outliers that were extreme, or that otherwise drastically altered the assumptions of linearity, were removed from the data set. In all data sets, 95% confidence intervals were used to test significance of differences in soil moistures between start and end of summer, between snow regimes, and between inside and outside OTCs. Emmeans package was used to extract the estimated marginal means, which were then graphed using ggplot2.

3.3.3 Tea bag decomposition (% mass loss) analysis

The primary interests of this experiment were to measure the decomposition, expressed in mass loss (%), of green tea and rooibos tea at three snow depths (*Ambient, Medium*, and *Deep*). Mass loss was calculated as follows:

Mass loss (%) =
$$\frac{\text{tea mass}_{initial} - \text{tea mass}_{final}}{\text{tea mass}_{initial}} \times 100$$

The mass loss from drying the tea bags at 60°C for 48hrs was calibrating using the recorded value of mass loss due to drying measured before the tea bags were buried. The corrected value is based on one individual tea bag from each box of tea that corresponds to each buried tea bag.

To account for differences in tea plant material, and seasonal decomposition differences, green tea and rooibos tea bags were kept separate in the analysis, and decomposition was measured for annual, winter, and summer (OTC and non-OTC) samples. The complete data set was divided into the following sections: decomposition of green tea: annual, winter, summer v1 (tea bags buried in the summer season), summer v2 (annual decomposition-winter decomposition), summer inside

OTCs; and decomposition of rooibos tea: annual, winter, summer v1, summer v2, and summer inside OTCs. Summer v2 decomposition was calculated as a comparison to summer v1 data as a result of high mass loss found in the summer v1 data. Any negative decomposition values calculated when subtracting winter tea bags from annual tea bags were treated as zeros, since winter decomposition contributed to the majority of annual decomposition in some cases.

Analyses were conducted in the statistical program R Studio, version 1.0.153 (RStudio, Inc 2009-2016). Decomposition in each snow depth regime was quantified using linear modelling, setting mass loss as a response and snow regimes (A, M, D) as predictor variables. For all models, we implemented fixed effects only, using the lm function, since insufficient replication led to singular fit, when implementing nested random design variables, such as blocks and the location of fences. Linear modelling (Im function) was also used to analyse the effect of OTCs. Normality was visually determined by using a box plot to give an initial overview of the data, and a Q-Q plot of model residuals. Residuals were also visually checked for homoscedasticity. Outliers were kept or removed based on their "mildness" or "extremeness" as an attempt to determine their ecological importance using the quartile method described in Benhadi-Marín (2018). Outliers that were extreme, or otherwise drastically altered the assumptions of linearity, were removed from the data set. For the OTC summer decomposition data sets, a likelihood ratio test was used to assess if an additive or a multiplicative model to test interaction significance. An Analysis of Variance (ANOVA), in combination with AIC values for both models (snow enhancement and OTC treatment as additive or multiplicative predictors) were compared to determined the significance of an interaction between the effect of OTCs and snow regime on mass loss, resulting in the use of an additive model (% loss ~ snow treatment + OTC treatment) for both green and rooibos tea. In all data sets, confidence intervals were used to determine statistical significance. We termed effect sizes with 95% confidence intervals not overlapping zero as statistically significant. Model estimated means were extracted using emmeans package. Ggplot2 was then used to visualize the results.

4 Results

4.1 Temperature

4.1.1 Winter

For a period of approximately 180 days during the winter season, soil temperatures were higher in *Deep* compared to both *Ambient* and *Medium* (see Figure 5). *Medium* was also higher than Ambient for approximately 160 days (Figure 5). Daily minimum temperatures (Figure A1 in Appendix A) were buffered in both *Deep* and *Medium* during winter, although temperatures were sometimes higher in Deep than Medium and vice versa throughout the winter. Minimum daily temperatures in *Deep* were more stable throughout the wintertime compared to both *Medium* and Ambient regimes. Minimum daily temperatures in Ambient snow regime reached -17.5°C in the coldest period of winter (March and early April), whilst the Medium regime reached -10°C and -7.5°C in *Deep* during this same cold period. Maximum daily temperatures (Figure A2) were, on average, more stochastic in Ambient compared to Medium and Deep, and reached warmest temperatures in *Ambient*, followed by *Deep*, with coolest daily maximum temperatures in *Medium*. Greater variation between minimum and maximum daily temperatures was also observed in Ambient compared to *Medium* and *Deep* (Figure 6). Below 0°C average temperatures (Figure A3) were the most extreme in Ambient, followed by Medium, whilst the most mild below 0°C temperatures were observed in *Deep* snow regimes, suggesting a buffering effect in *Deep*. Warmer average temperatures were reached first in the *Ambient* regime during the period of spring melt-out (Figure 7), from -8°C to slightly above 0°C between doy=125 (5 May) and doy=135 (15 May). Medium reached above 0° C temperatures a few days after that (-6°C to above 0°C between doy=125 and doy=142 (22 May)), while *Deep* reached average daily temperatures above 0°C only after doy=150 (30 May). Snow accumulation likely began around doy=315 (11 November) in 2017, when the temperatures begin to stabilize.



Figure 5. Annual daily mean temperatures outside OTCs (48 loggers) for each snow regime (A, M, D) using Tiny tag loggers planted in September 2017 and removed August 2018.



Figure 6. Annual daily minimum and maximum temperatures outside OTCs for each snow regime.



Figure 7. Spring daily mean temperatures outside of OTCs for each snow regime.

4.1.2 Summer

An increase in mean temperature was observed in all snow regimes from doy=120 until doy=190, where they remained relatively stable until approximately doy=210 before temperatures then began to drop in all regimes (Figure 5). Daily mean temperatures throughout the summer period are relatively similar between snow regimes, although warmer temperatures are commonly observed in Ambient and Medium, while Deep had lower mean daily temperatures (Figure 8). Maximum daily temperatures (Figure 9) throughout snow regimes fluctuate throughout the summer period. There was no noticeable regularity of highest consistent temperatures between snow regimes. However, minimum daily temperatures (Figure 9) were more stable for all three of the snow regimes. Minimum daily temperatures were continually lowest in *Deep* snow regime plots, and highest in Medium snow regime. Snow regimes with enhanced temperatures using OTCs were consistently higher than all snow regimes without OTCs (Figure 10). There was more temperature variance between Deep, Medium, and Ambient snow regimes in non-OTC plots than inside OTC plots. In contrast to the winter period, below 0°C temperatures (Figure A3) during the summertime were more extreme in *Deep* snow regimes, and also cooler in *Ambient* snow regimes. Below 0°C temperatures in *Medium* were consistently near 0°C throughout the summer. Relatively similar maximum daily temperatures were observed when comparing OTC and non-OTC plots within all snow regimes (Figure 11). However, minimum daily temperatures were never below 0°C inside OTCs, while below 0°C was observed outside of OTCs, and there was very little noticeable temperature difference between snow regimes inside OTCs. An example of hourly fluctuations of aggregated temperature for a 24 hour period is given during summer (doy=200) to highlight the difference of temperature variance outside of OTCs (Figure A4) and inside of OTCs (Figure A5). Figure A4 displays mean, minimum, and maximum hourly temperatures, while Figure A5 displays only minimum and maximum hourly temperatures.



Figure 8. Summer (doy=172 to doy=239) daily mean temperatures outside OTCs for all snow regimes.



Figure 9. Summer daily minimum and maximum temperatures outside OTCs.



Figure 10. Summer daily mean temperatures inside OTCs compared to outside OTCs for all snow regimes.



Figure 11. Summer daily minimum and maximum temperatures inside OTCs for all snow regimes.

4.2 Soil Moisture

4.2.1 June



Figure 12. Soil moisture measurements in June, inside and outside OTCs, for all snow regimes (58 observations) using estimated means and their 95% confidence intervals.

Soil moisture measurements were taken inside and outside of OTCs in June, immediately after the erection of the OTCs. No significant difference was found between soil moisture in the three snow regimes inside compared to outside of OTCs, and therefore June soil moisture values are an amalgamate of inside and outside OTCs (see Figure 12). No significant difference between *Medium* and *Ambient* was observed (lower limit=-127.39, mean=41.13, upper limit=209.65V), nor in *Deep* compared to *Ambient* (lower limit=-76.87 mean=87.15V, upper limit=251.17V).

4.2.2 September



Figure 13. Soil moisture measurements for September (a) inside and outside OTCs between snow regimes (58 observations) and (b) inside versus outside of OTCs (58 observations) showing estimated means and their 95% confidence intervals. No interaction was found between the effect of snow regime and the effect of OTC.

Soil moisture measurements were taken again inside and outside of OTCs at the beginning of September, marking the end of the summer season. An interaction was found between snow regime treatments and the effect of OTCs on soil moisture for the September measurements (Figure 13a). Page 18 of 47 The effect of OTCs on the *Medium* regime was not significant (lower limit=-189.56V, mean= 99.40, upper limit=388.36V), as was the effect of OTCs on *Deep* (lower limit=-209.16V, mean=72.10V, upper limit=353.36V). Soil moisture inside OTCs was not significantly different to outside of OTCs (lower limit=-284.28V, mean= -85.40V, upper limit=113.48V) (see Figure 13b). Figure A6a) shows no significant difference in soil moisture between *Medium* outside of OTCs compared to *Ambient* (lower limit=-198.21V, mean=-7.40V, upper limit=183.41V). No significant difference in soil moisture between *defium* outside of OTCs compared to *Ambient* (lower limit=279.42V). Inside of OTCs (Figure A6b), no significant difference in soil moisture was observed in *Medium* (lower limit=-134.30V, mean=92.00V, upper limit=318.29V), and no significant difference in soil moisture in *Deep* compared to *Ambient* (lower limit=-54.46V, mean=165.80V, upper limit=386.06V) was found. Figure A7 and A8 compare soil moisture differences between June and September, inside and outside of OTCs, respectively.

4.3 Annual Decomposition



Figure 14. Annual mass loss (%) of green tea between snow depth regimes (A, M, D) (28 observations) buried in September 2017 and removed in late August 2018 showing estimated means and their 95% confidence intervals.

Figure 15. Annual mass loss (%) of rooibos tea between snow depth regimes (A, M, D) (30 observations) buried in September 2017 and removed in late August 2018 showing estimated means and their 95% confidence intervals.

Green tea

Decomposition (% mass loss) in green tea bags from September 2017 to August 2018 (355 days in soil) (see Figure 14) was higher in the *Medium* snow regime than in the *Ambient* snow regime by a mean of 3.69 increase in mass loss (%). Confidence intervals do not suggest significance (lower limit=-0.53, mean=3.69, upper limit=7.91), however the overlap with zero is small, suggesting a potential biologically significant increase in mass loss. Decomposition was not significantly lower in *Deep* than in *Ambient* (lower limit=-6.58, mean=-2.22, upper limit=2.15).

Rooibos tea

Decomposition in rooibos tea bags from September 2017 to August 2018 (see Figure 15) was not significantly different in the *Medium* snow regime than in *Ambient* (lower limit=-2.09, mean=-0.71, upper limit=0.66). Decomposition was not significantly different in *Deep* compared to both the *Medium* and *Ambient* snow regimes (lower limit=-2.61, mean=-1.27, upper limit=0.07). However, the confidence interval overlap of zero is very small, possibly suggesting biological significance.



4.4 Winter Decomposition

Figure 16. Winter mass loss (%) of green tea between snow depth regimes (A, M, D) (30 observations) buried in September 2017 and removed in late June 2018 using estimated means and their 95% confidence intervals.

Figure 17. Winter mass loss (%) of rooibos tea between snow depth regimes (A, M, D) (32 observations) buried in September 2017 and removed in late June 2018 using estimated means and their 95% confidence intervals.

Green tea

Winter decomposition (September 2017-June 2018, 286 days in soil) of green tea bags (see Figure 16) was not significantly different in *Medium* compared to *Ambient* (lower limit=-1.54, mean=1.45, upper limit=4.44). Tea bags buried in *Deep* did not decrease significantly in compared to *Ambient* plots either (lower limit=-3.99, mean=-1.01, upper limit=1.98).

Rooibos tea

Winter decomposition of rooibos tea bags (see Figure 17) was similar in *Medium* compared to *Ambient* (lower limit=-1.32, mean=-0.09, upper limit=1.14), and decomposition in *Deep* was almost the same as *Ambient* (lower limit=-1.31, mean=-0.11, upper limit=1.09). Little change in decomposition between snow regimes was observed in rooibos tea bags during the wintertime.

4.5 Summer Decomposition, non-OTC



4.5.1 Summer Version 1 (tea bags buried during summer season)

Figure 18. Summer mass loss (%) of green tea between snow depth regimes (A, M, D) (30 observations) buried in June 2018 and removed in late August 2018 using estimated means and their 95% confidence intervals.

Figure 19. Summer mass loss (%) of rooibos tea between snow depth regimes (A, M, D) (32 observations) buried in June 2018 and removed in late August 2018 using estimated means and their 95% confidence intervals.

Green Tea

Green tea bags buried during the summer season (Figure 18) from June 2018-August 2018 (69 days in soil) showed a significant increase of decomposition in *Medium* compared to *Ambient* (lower limit=0.48, mean=3.61, upper limit=6.72). No significant difference of decomposition in the *Deep* regime was observed (lower limit=-1.45, mean=1.57, upper limit=4.61).

Rooibos tea

Rooibos tea bags buried during the summer season (see Figure 19) in *Medium* showed no significant increase in decomposition compared to *Ambient* (lower limit=-1.44, mean=0.54, upper limit=2.51). Bag buried in under the *Deep* regime showed a similar decomposition compared to *Ambient* (lower limit=-2.06, mean=-0.13, upper limit=1.80).



4.5.2 Summer Version 2 (annual tea bags - winter tea bags)

Figure 20. Calculated summer mass loss (%) of green tea between snow depth regimes (A, M, D) (28 observations) for annual decomposition – winter decomposition (summer version 2 decomposition) using estimated means and their 95% confidence intervals.

Figure 21. Calculated summer mass loss (%) of rooibos tea between snow depth regimes (A, M, D) (32 observations) for annual decomposition – winter decomposition (summer version 2 decomposition) using estimated means and their 95% confidence intervals.

Green tea

The calculated difference of winter green tea bags subtracted from annual green tea bags (see Figure 20) resulted in an obvious, but statistically non-significant increase in decomposition in the *Medium* regime compared to *Ambient* (lower limit=-0.60, mean=3.62, upper limit=7.83). Green tea in *Deep* showed a similar decomposition compared to *Ambient* (lower limit=-4.98, mean=-0.62, upper limit=3.73).

Rooibos tea

The calculated difference of winter rooibos tea bags subtracted from annual rooibos tea bags (see Figure 21) did not result in a significant difference between *Medium* compared to *Ambient* (lower limit=-1.89, mean=-0.01, upper limit=1.87). A non-significant decrease in decomposition was also observed in *Deep* compared to *Ambient* (lower limit=-2.54, mean=-0.71, upper limit=1.13).



4.6 Summer Decomposition and the OTC effect

Figure 22. Summer mass loss (%) of green tea inside of OTCs between snow depth regimes (A, M, D) (26 observations) buried June 2018 to August 2018 using estimated means and their 95% confidence intervals.

Figure 23. Summer mass loss (%) of green tea inside and outside of OTCs between snow depth regimes (A, M, D) (26 observations) buried June 2018 to August 2018 using estimated means and their 95% confidence intervals.

Figure 24. Summer mass loss (%) of green tea between OTC treatments (where "yes" signifies OTC presence and "no" signifies OTC absence) (58 observations) for all snow treatments using estimated means and their 95% confidence intervals.



Figure 25. Summer mass loss (%) of rooibos tea inside of OTCs between snow depth regimes (A, M, D) (26 observations) buried June 2018 to August 2018 using estimated means and their 95% confidence intervals.

Figure 26. Summer mass loss (%) of rooibos tea inside and outside of OTCs between snow depth regimes (A, M, D) (26 observations) buried June 2018 to August 2018 using estimated means and their 95% confidence intervals.

Figure 27. Summer mass loss (%) of rooibos tea between OTC treatments (where "yes" signifies OTC presence and "no" signifies OTC absence) (58 observations) for all snow treatments using estimated means and their 95% confidence intervals.

Green tea

No significant interaction was found between snow regime treatments and open top chambers for green tea bags buried during the summer, confirming the effect of snow regime to be independent to the effect from OTCs. Green tea bags buried inside of OTCs (see Figure 22) for a summer period (June 2018-August 2018, 66 days in soil) showed no significant difference in decomposition in both *Deep* (lower limit=-1.01, mean=2.15, upper limit=5.31) and *Medium* compared to *Ambient* (lower limit=-1.13, mean=2.20, upper limit=5.53). Green tea bags buried for a summer period from June 2018-August 2018 (66 days in soil) inside and outside of OTCs (see Figure 23) showed no significant difference in decomposition in *Medium* compared to *Ambient* (lower limit=-0.85, mean=1.79, upper limit=4.43), however the small overlap of 0 for the confidence intervals may suggest ecological significance. A significant increase in decomposition for inside and outside of OTCs was found in *Deep* compared to *Ambient* (lower limit=0.26, mean=2.81, upper limit=5.36). No significant difference in decomposition (see Figure 24) was found in bags buried inside OTCs compared to outside of OTCs (lower limit=-3.17, mean=-1.05, upper limit=1.06).

Rooibos tea

No significant interaction was found between snow regime treatments and open top chambers for rooibos tea bags buried during the summer, confirming the effect of snow treatment to be independent to the effect of OTCs. Rooibos tea bags buried in *Medium* inside of OTCs (see Figure 25) for a summer period (66 days in soil) showed no significant difference in decomposition compared to *Ambient* (lower limit=-1.53, mean=0.64, upper limit=2.81). No significant difference in decomposition was observed in *Deep* (lower limit=-1.00, mean=1.12, upper limit=3.23). In combined decomposition values for rooibos tea bags buried during the summer inside and outside of OTCs (see Figure 26), no significant difference in decomposition was observed in *Deep* either, although this slight increase in *Medium* compared to *Ambient* (lower limit=-1.24, mean=0.25, upper limit=1.74. No significant difference in decomposition was observed in *Deep* either, although this slight increase in *Deep* was smaller than the increase in *Medium* (lower limit=-1.33, mean=0.12, upper limit=1.58). A significant decrease in decomposition inside OTCs compared to outside OTCs (see Figure 27) was found (lower limit=-3.81, mean=-2.60, upper limit=-1.39).

4.7 Decomposition overview

A complete table summarizing the effect sizes and the corresponding confidence intervals for green and rooibos tea bags in each snow depth regime can be found in in Table 1 below.

	Effect st	trength Mee Depth	dium Snow	Effect	strengths De Depth	eep Snow	Interaction-effect st		trength of OTC
	Lower Limit	Mean	Upper Limit	Lower Limit	Mean	Upper Limit	Lower Limit	Mean	Upper Limit
Green Tea Annual	-0.53	3.69	7.91	-6.58	-2.22	2.15		N/A	
Green Tea Winter	-1.54	1.45	4.44	-3.99	-1.01	1.98		N/A	
Green Tea Summer v.1	0.48	3.61	6.72	-1.45	1.57	4.61		N/A	
Green Tea Summer v.2	-0.60	3.62	7.83	-4.98	-0.62	3.73		N/A	
Green Tea OTC only	-1.13	2.20	5.53	-1.01	2.15	5.31		N/A	
Green Tea OTC and non-OTC Summer	-0.85	1.79	4.43	0.26	2.81	5.36	-3.17	-1.05	1.06
Rooibos Tea Annual	-2.09	-0.71	0.66	-2.61	-1.27	0.07		N/A	
Rooibos Tea Winter	-1.32	-0.09	1.14	-1.31	-0.11	1.09		N/A	
Rooibos Tea Summer v.1	-1.44	0.54	2.51	-2.06	-0.13	1.80		N/A	
Rooibos Tea Summer v.2	-1.89	-0.01	1.87	-2.54	-0.71	1.13		N/A	
Rooibos Tea OTC only	-1.53	0.64	2.81	-1.00	1.12	3.23		N/A	
Rooibos Tea OTC and non- OTC Summer	-1.24	0.25	1.74	-1.33	0.12	1.58	-3.81	-2.60	-1.39

Table 1. Effect sizes and corresponding 95% confidence intervals for green and rooibos tea, for all periods of decomposition (annual, winter, and summer (V.1 and V.2)), inside and outside of OTCs, in *Medium* and *Deep* snow regimes compared to *Ambient*. Bolded values signify significance, and italicized values signify near significance.

5 Discussion

The aim of this experiment was to assess the effect of varying snow depths on annual, winter, and summer decomposition of rooibos and green tea plant material, combined with the effects of enhanced summer temperatures on summer decomposition. I hypothesized that both types of tea plant material would decompose most rapidly under the *Deep* (D) snow regime, slightly less rapidly under the *Medium* snow regime (M), and most slowly in the *Ambient* (A) snow regime. I also hypothesized this would be the case in both winter and summer, due to the insulation effects of snow cover during the wintertime, and increased soil moisture in the summertime from the snow cover during the winter period. I expected that increased summer temperatures inside OTCs would enhance summer decomposition. However, after analysis of the results, it is clear that the processes of decomposition are much more complex than their dependency on winter soil insulation and consequent summer soil moisture. The following sections will discuss in detail the processes affecting decomposition for the two types of tea in my experiment.

5.1 355 days of decomposition

5.1.1 Winter decomposition and the effects of fall-winter-spring temperature

Temperature is a main driver of soil microbial processes, and studies have shown that higher temperatures increase decomposition rates of SOM (Walz et al., 2017). In this study, we did observe increased winter temperature with increased snow depth, where *Deep* had the warmest winter temperatures, followed by *Medium*, and the coldest winter temperatures in *Ambient* (see Figure 5). The temperature loggers used in this experiment give clear results that the different depths of snow cover accumulation in the wintertime do affect soil temperatures throughout all seasons, but the results for decomposition were unexpected after low decomposition was recorded in the *Deep* regime during the winter period (September to June). For green tea bags, we observed the highest winter decomposition in *Medium* compared to *Ambient*, and the lowest winter decomposition in *Deep* compared to *Ambient* (see Figure 16), which was the opposite to what was hypothesized considering the increased winter temperature in *Deep*. Although the difference between snow regimes for rooibos tea bags was small, we observed the lowest winter decomposition in *Deep*, and highest in *Ambient* (see Figure 17). The effect of temperature during the individual periods of fall, winter, and spring seasons therefore likely play an important role on our cumulative winter decomposition values.

Before the onset of winter, during the fall period when the tea bags were first buried, lowest minimum temperatures were observed in *Deep* and *Ambient* regimes (see Figure A1). This is a very important time for decomposition, as the labile material from dead arctic plant species is being quickly broken down, and the newly introduced tea bags are readily available for microbes. This addition of new tealeaf material in September may cause a shift in preference of microbes, otherwise demonstrating declining microbial activity, to new labile plant material. Warmer temperatures during this period could therefore have particular importance on decomposition, consequently influencing the overall winter decomposition. There was little discrepancy in the variation of mean and maximum daily temperatures among snow regimes during this period, which places emphasis on the importance of the drops in temperature experienced by each snow treatment plot. The *Medium* snow regime displays the warmest temperature during fall (see Figure A2), and may contribute an explanation as to why *Medium* regimes displayed largest winter decomposition in green tea bags, and second largest winter decomposition in rooibos tea bags.

Once the snow began to accumulate, winter temperature loggers show a buffering effect of increased snow depth in the *Deep* snow regime (see Figure 5). Temperatures inside the *Deep* are more stable, and warmer than *Medium* and *Ambient* regimes, and the same effects have been observed in other studies (Morgner et al., 2010; Mörsdorf et al., 2019; Semenchuk et al., 2015). The buffering effect can still be seen in *Medium*, while fluctuating temperature swings and very cold periods are clearly observed in *Ambient*. Theoretically, this should imply higher decomposition rates in the *Deep* snow regimes, due to higher winter temperatures. However, data for both green and rooibos teabags consistently indicates otherwise in this study. Interestingly, it was observed that, at several points during the winter, the *Medium* regime had daily minimum temperatures that were several degrees warmer than in *Deep. Ambient* plots were consistently colder throughout the snow-covered winter period. The cold winter temperatures in *Ambient*, paired with high decomposition for rooibos teabags in both the annual and winter decomposition periods might suggest that decomposition is greatly influenced by factors other than winter temperature.

At the onset of spring, in agreement with other studies, soil temperatures gradually increase, and then drastically peak to the zero curtain at the time of snowmelt (Seastedt et al., 2001). This snow melt phase is very important for soil microbial activity. As temperatures begin to increase abruptly, osmotic pressure in the cells of the winter microbes increases, potentially resulting in cell burst and microorganism death within the soil system (Lipson et al., 1999; Schimel et al., 2007). This influx of cell material contains dissolved organic nitrogen and carbon, which, paired with the favourable rising temperatures for the surviving summer microbes, provides nutrients to increase

nutrient cycling activity (Gavazov, 2010). The highest N availability in soil is often found right after snowmelt (Mörsdorf et al., 2019), making the timing of snow melt for each snow regime treatment very important for decomposition.

When the snow begins to melt, the temperature loggers display a rapid increase in warming in the *Ambient* regime, closely followed by *Medium* (see Figure 7). However, the delay in snowmelt is seen clearly in *Deep*, where above zero temperatures are delayed for two weeks after the warming of *Ambient* and *Medium*.

Due to the timing of tea bag removal for the winter data set and the burying of new tea bags for the summer data set, a potentially important sampling bias occurred. Tea bags were removed and buried in each snow regime at the same time (19 June, 2018, doy=170), regardless of the onset of snow melt. Consequently, the winter period is potentially wrongly defined in both the *Ambient* (snow melts around 15 May) and *Medium* (snow melts around 22 May) snow regimes, as they include the unique nutrient influx that comes with the onset of spring. This may present higher decomposition in my winter data set than should be expressed. The corresponding summer data sets are likely to be wrongly defined in *Medium* and *Ambient* as well, as they do not include the snowmelt nutrient influx. The *Deep* regime in the winter data set may not have included the important event of snowmelt (snow melts around 30 May), which was likely included in the summer data set instead (beginning 19 June). This may explain the lower values for decomposition in the winter data set for *Deep* for both green and rooibos tea.

The warming effect of increased snow depth during the wintertime did not have as much influence on decomposition as I had originally suspected, either due to sampling bias, or possibly due to other factors affecting decomposition. There are plant community differences, and thus litter quality differences, in the snow regimes of this experiment (Cooper et al., in review). It has also been found that the decomposition of SOM in temperatures close to freezing may not respond as strongly to changes in temperature (Walz et al., 2017). This might suggest that, despite the fluctuating low temperatures in the *Ambient* regime during wintertime, below zero temperatures are observed in all plots, and the influence of snow depth warming on decomposition may not be as important as the SOM quality created by the corresponding above-ground plant communities. This could explain why the microbial communities in the *Ambient* plots were found to be more productive than in the *Deep* regime, despite the warmer temperatures. Because other studies have clearly demonstrated the important influence of soil temperatures on winter nutrient cycling, the importance of considering potential microbial community shifts in long term studies is emphasized (Mikan et al., 2002).

5.1.2 Summer decomposition without open top chambers

Lower below zero temperatures are observed throughout the summertime in the *Deep* regime, while the warmest below zero temperatures are observed in the *Medium* plots (see Figure A3). Highest daily minimum temperatures are shown in *Medium*, followed by slightly less warm temperatures in *Ambient*, and coldest daily minimum temperatures in *Deep* (see Figure 9). Furthermore, higher daily maximum temperatures are observed in *Medium* and *Ambient*, while the lowest daily maximum temperatures are seen in *Deep* snow plots. Both green tea and rooibos tea had highest mass loss rates in *Medium* during the summer period (see Figure 18 and 19), which may be partly explained by these temperature findings.

The combination of buffered winter soil temperatures, relatively warmer summer temperatures, and balanced soil moisture content (not too dry, not too moist) helps to explain the result of green and rooibos tea decomposition in the summer. Furthermore, the shrub community in *Medium*, although not significant, was reported to be slightly less than in *Ambient* (Cooper et al., in review) which might point to slightly less shading effect on the soil surface from plant cover in the summer, giving rise to higher summer temperatures in this regime. Leaching loss and the uptake of nutrients by plants create a considerable nutrient competition for soil microbes during the growing season (Schimel et al., 2004). In the *Deep* regime, where previous studies have shown high N concentrations in these plots, plant competition for nutrient uptake may influence the lower decomposition rates found in this study (Mörsdorf et al., 2019; Semenchuk et al., 2015).

The summer decomposition data set of this experiment, however, is not a great indication for what truly may be happening in arctic soil ecosystems. Tea bags that were buried for the summer data set were inserted directly into the same holes of the previously removed winter tea bags, which may have had an effect on the decomposition if the microhabitat was influenced by the presence of tea bags in the winter. Furthermore, due to the experimental design bias of removing tea bags at the same time of year, regardless of onset of snowmelt, summer decomposition is possibly overestimated in the *Deep* regime and under-estimated in the *Ambient* and *Medium* regimes, which makes the estimations of mass loss difficult to translate. Another experiment in an alpine dry meadow found that microbial biomass was highest in the cold season (fall-winter-spring), and decreased after snowmelt to have lower microbial biomass during the summer (Lipson et al., 1999). We found higher decomposition during the summertime than during the winter period despite the shorter incubation period, which suggests that planting the summer tea bags in the same holes as the winter tea bags may have created a bias in decomposition measurements. Although I would expect initial

decomposition to be higher in the summer than in the winter due to increased temperature and moisture availability, without tea bags planted in new holes, it's difficult to know if temperature and moisture are the sole causes of increased decomposition.

The summer Version 2 mass loss was calculated in the attempt to compliment the tea bags buried for the summer period. However, this calculated mass loss using winter decomposition subtracted from annual decomposition misses a lot of important information, especially the initial rates of faster decomposition and the subsequent stabilizing period, making it very difficult to use as an explanation for the processes happening beneath the soil. This summer Version 2 data is therefore not used to draw any sort of conclusions.

5.1.3 Potential litter feedback influences on decomposition

Temperature has a direct effect on leaf litter mass loss, but can also indirectly cause shifts in plant community structure and indirectly affect soil organic carbon dynamics (Cornelissen et al., 2007; Walz et al., 2017). Decomposition does not only depend on the direct effect of temperature and moisture, but also greatly on the type of organic material being decomposed. While snow depth directly affects litter decomposition by changing winter soil temperatures and summer soil moisture and temperature, long-term studies, such as the SnoEco snow fence experiment, might not be as influenced by short-term winter temperature effects. Our results suggest that the litter type and the plant community composition of the snow depth treatments may play a much more important role than was originally considered, given the overall low decomposition found in the *Deep* regime for both green and rooibos tea, during winter (see Figures 16 and 17) and for the entire year (Figure 14 and 15). Changes in plant and microbial composition may have occurred after 13 years of manipulating snow accumulation, corresponding to the changes in plant community that have recently been found at our site (Cooper et al., in review). It is therefore extremely important to consider plant litter type and plant community composition to understand the results of this experiment. It has been repeatedly shown that plant functional group characteristics show distinctly different effects on soil nutrient availability due to differences in litter quality and the mechanisms of decomposition (Gavazov, 2010; Hobbie, 1992). It has also been found that the nature of available substrates has important control over microbial activity, where the effect of plant species type on decomposition rates is greatly determined by lignin (Baptist et al., 2010) which differs between shrubs and forbs.

A study in 2006 within the International Tundra Experiment found that warming temperatures across the tundra biome resulted in increased height and cover of deciduous shrubs and graminoids, while overall species diversity and evenness was observed to decrease (Walker et al., 2006). A possible feedback loop has been predicted wherein increased snow cover results in increased soil temperatures during the winter, supporting an expansion of shrubs due to increased nutrient cycling via increased winter microbial activity (Sturm et al., 2000, 2005). Contrary to the proposition by Sturm et al., very recently, plant community shifts have been observed behind the snow fences of Adventdalen, corresponding to manipulated snow depth (Cooper et al., in review). This trend towards "browning" in certain areas of the high Arctic may suggest that long term plant community changes vary regionally and locally across all latitudes, creating a mixture of greening and browning sites (Cooper et al., in review; Phoenix et al., 2016; Sturm et al., 2000). The nature of the changes in vegetation composition and dominant species presence could cause shifts in litter quality and decomposability, contributing to the release of carbon dioxide into the atmosphere (Cornelissen et al., 2007). However, it has also been observed that changes to plant communities with poorer litter quality will create a negative feedback loop in carbon emissions, as the ease at which microbes can decompose material decreases with lower leaf litter quality, which may be the case for increased shrub dominance (Cornelissen et al., 2007). The increases in mosses found in *Medium*, and decreases in living shrubs found in *Deep* will likely affect the microbial community beneath these regimes. The ability for thin layers of mosses to regulate their microenvironment might have positively affected the decomposition in *Medium*, resulting in increased decomposition of tea (see Figures 16, 18, 19) (Gornall et al., 2007). However, Gornall et al. (2007) also found that deeper layers of moss cover reduce soil temperature and moisture, which might suggest alternative plant community shifts in decomposition at our site if the depth of the moss layer increases.

5.1.4 Putting the pieces together: Annual decomposition

Annual decomposition values hold the highest ecological significance for this study. They include all the important phases of decomposition throughout the year, and were not influenced by sampling bias around the time of snowmelt. Distinct microbial communities have been discovered in the active layer of permafrost soils in Svalbard between winter and summer periods, and the annual data set of this experiment allows the transitions of microbial communities to be uninterrupted by the timing of sampling (Schostag et al., 2015). Considering summer moisture, increased winter and summer temperatures, and the influence of the plant community composition in *Medium*, the highest decomposition in the *Medium* regime can be explained, where decomposition is strongly influenced by temperature, moisture, litter quality and microbial composition. The unexpected high decomposition in *Ambient* might suggest that the influence of plant community composition under this snow regime, which is dominated by shrubs, is stronger than the effect of winter warming due to

snow cover. Although the *Ambient* regime has colder winter soil temperatures (see Figure 5), the microbial activity here seems to be higher than the warmed *Deep* regimes. Furthermore, the observation that rooibos tea bags decomposed the most in *Ambient* may suggests that this recalcitrant litter was highly compatible with the local microbial community, where the litter substrate of shrubs is slightly more recalcitrant by nature (Dorrepaal et al., 2005; Veen et al., 2015).

Explaining decomposition rates for both green and rooibos tea in the *Deep* regime is more difficult than simply considering increased winter temperatures and increased summer moisture. The complex processes of decomposition in arctic soils that have been influenced by long term studies, as has been seen at the snow fences in Adventdalen, has surely influenced the decomposition we observed in *Deep*. We found overall lower rates of decomposition in the summer *Deep* regime, despite the potential over estimation due to the inclusion of snowmelt in this period. This suggests that decomposition rates in both summer and winter are affected by the long-term shift in plant and microbial community of *Deep*, and may now simply express lower microbial activity. The *Deep* regime at this study site is now less dominated by living vascular plants, with a lower shrub cover (Cooper et al., in review) and perhaps an overall changed microbial community. Changes in fungi communities due to snow depth manipulation was found in 2016 at our site, and may give reason to believe that further microbiological changes have since occurred (Mundra et al., 2016). Lower summer temperatures, likely influence by increased moisture, found in our *Deep* regime may therefore be responsible for overall decrease in microbial activity during the normally productive summer period. Without microbial samples, however, it's difficult to make a complete conclusion.

Figure 28 below illustrates the potential cyclic effects of increased snow to varying depths (*Deep* or *Medium*).



Figure 28. Schematic diagram for increased winter snow depth to *Medium* or *Deep*, compared to *Ambient*, on annual decomposition (not including the effect of OTCs) at our snow fence site, based on the discussion so far. Arrows indicate direction of flow; red dashed boxes represent the physical responses of increased snow depth during winter; blue boxes represent long term composition changes that occurred at the Adventdalen snow fences before I began my study; yellow dashed boxes represent decomposition trends I observed in my study; and black boxes represent theoretical responses that I speculate to occur.

5.2 OTC effects: the importance of moisture and lack of OTC interactions with enhanced snow depths

Snow regime effect and temperature increase in summer due to the addition of OTCs acted independently from each other, which is aligned with the recent findings of Mörsdorf et al. (2019). Decomposition was significantly slowed inside OTCs in rooibos tea bags, and nearly significantly delayed in green tea bags as well. Other studies have shown similar results, where polythene tents were found to increase air, litter, and soil temperature, but reduce litter and soil moisture contents, having an overall effect of reducing decomposition (Robinson et al., 2017).

Soil moisture decreased from June to September, inside and outside of OTCs, in all snow regimes, indicating a drying period during arctic summers. Soil moisture levels remained the highest

in the *Deep* regime by the end of summer both inside and outside of OTCs, which demonstrates the important influence changes in snow regimes have on summer soil conditions. In September soil moisture measurements, an interaction was found between the effect of snow regime and OTCs, indicating that the effect of OTCs is combined with the effect of the winter snow depth to influence soil moisture measurements at the end of the summer. Increased moisture in *Deep* compared to *Ambient* and *Medium* regimes inside and outside of OTCs is a likely influencer in the decreased temperatures we observed in these snow regimes throughout the summer season (Bunnell et al., 1977). Decreased soil moisture inside of OTCs can be a good explanation of observed lower decomposition inside of OTCs compared to outside of OTCs.

Considering temperatures inside OTCs, we found warmer temperatures in all snow regimes compared to temperatures outside of OTCs (see Figure 10), creating a very similar temperature profile among the three snow regimes inside OTCs. However, on a few occasions throughout the summer, the highest temperatures were reached in the *Medium* regime, while the lowest were reached in the *Deep* regime, although the differences were minimal. Because the temperatures can be attributed to the heat of direct sunlight on the sensors. Despite warmer daily minimum temperatures inside (see Figure 11) of OTCs than outside (see Figure A3), decomposition was still lower inside the OTCs for both green tea and rooibos tea (see Figure 24, 27). This suggests the roles of soil moisture inside OTCs are more likely to explain the results for lower decomposition inside of OTCs. Other experiments have also shown that litter decomposition was significantly delayed inside tent-like warming treatments, likely due to reduced litter moisture and reduced soil moisture (Aerts, 2006; Robinson et al., 2017).

Future soil moisture conditions have been difficult to predict, and it is uncertain whether climate change will bring drier and warmer summer conditions to the Arctic, or if increased summer precipitation is also a possibility (Kattsov et al., 2007). If increased precipitation during the summer time becomes the trend in our changing climate, the use of OTCs might not be an accurate proxy for predicting future soil microbial responses to the addition of leaf litter.

5.3 Difference between rooibos and green tea

The results of this experiment support previous studies by visually demonstrating differences in decomposition between green tea and rooibos tea. Although this was not tested for significance, green tea consistently showed higher decomposition than rooibos tea, suggesting a higher content of more labile materials in green tea compared to rooibos tea.

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In a laboratory experiment that set the foundation for the Tea Bag Index, green tea bags were characterized by a significantly lower C:N ratio of 12.23, while rooibos tea bags had a C:N ratio of 42.87 (where lower C:N ratio indicates easier decomposition) (Keuskamp et al., 2013). Green tea also had a higher hydrolysable fraction (H) of 0.84g g⁻¹ than rooibos, 0.55g g⁻¹, where H is assumed to be rapidly decomposable (Keuskamp et al., 2013). After a period of being buried in soil, the initial decomposition of green tea was rapid, levelling off between 40-60 days, while the initial decomposition of rooibos tea was much slower, only levelling off towards the end of the laboratory experiment (130 days). In experiments involving indigenous plant litter, forbs are found to decompose consistently more quickly than grasses or sedges, while herbaceous growth forms consistently decompose more quickly than shrub growth forms (Cornelissen et al., 2007). These similarities in patterns support the use of green tea, acting as a herbaceous plant, and rooibos tea, acting more as a shrub growth form, as a means to draw conclusions in decomposition rates of Arctic ecosystems.

The differences in results between rooibos and green tea decomposition under different snow depth treatments suggests there may be an effect of litter composition and the compatibility it has with a well-established microbial community. A study by Manzoni et al. (2008) suggests that decomposers can lower their carbon-use efficiency in order to maximize plant residues with low initial nitrogen concentration, providing evidence for a shift in microbial mineralisation processes depending on the above ground plant community (Manzoni et al., 2008). Perhaps this is occurring in the regimes that are more heavily dominated by recalcitrant shrubs. Other studies have shown that the "home-field advantage" hypothesis applies to plant litter decomposition, where leaf litter decomposes faster in soils from where the plant originates (Veen et al., 2015). In the case of tea leaf litter, while its place of origin lies far away from the high Arctic, we can still argue that green tea plant material resembles a herbaceous forb species, while rooibos tea resembles a more recalcitrant shrub-like species. Paired with the litter type, the dominant plant species associated with the specific snow regime (where the *Ambient* regime has a more shrub dominated plant community, the *Deep* regime has a significant decrease in living shrubs, and the *Medium* regime is somewhere in between with increased bryophyte communities (Cooper et al., in review), may have influenced the microbial communities below the soil, and therefore their interaction with the specific types of tea. Shifts in plant community composition and the inherit amount and quality of fresh and labile organic matter from the surface may also stimulate microbial activity and enhance or hinder the decomposition of older SOM (Fontaine et al., 2007).

The home field advantage theory, in part, may explain why decomposition of green tea and rooibos tea in the different snow depth regimes differed. The recalcitrant rooibos leaf material may have functioned better as a decomposable material in *Ambient*, which is dominated by recalcitrant material, compared to the green tea herbaceous leaf litter. These differences in response to snow regime treatment solidify the importance of species composition and the influence a changing climate may have on this ecosystem. After years of active snow fences in Adventdalen, it's likely that decomposition processes are adapting to changes in plant community, and that microbiological activity is now lower in the *Deep* regime.

5.4 Comparison of decomposition values to other Arctic litter studies

Although there are no known published studies that mimic this one, there are several litter (both tea and indigenous plant species) studies in the Arctic to which the decomposition values in this experiment can be compared. Djukic et al. (2018) created a global biome tea bag decomposition study, in which 200 tea bags were used to assess mass loss in the Arctic compared to other biomes around the world. Sample sites were located in Canada (Nunavut, Bylot Island, Umiujaq), Greenland, Russia (Cherskiy) and Sweden (Latnjajaure). This study took place from June to August, aligning with the summer period of my study. The mass loss for rooibos tea bags in the study of Djukic et al. (~25%) were close, but slightly lower, to my rooibos decomposition values, but the values for green tea were slightly higher (55-75%) (Djukic et al., 2018). This may be due to variations in temperatures and microbial composition between sites. However, the standard error bars for the green tea bags in Djukic's study were large, and may suggest that my green tea summer decomposition values could still be comparable.

A study in 2003 in Abisko, Sweden, showed mass loss in dwarf shrub species to be between 25 and 60% (depending on species) 35% for *Bistorta vivipara* and 55% for *Rubus chamaemorus* in a life form group experiment in outdoor litter beds over one year (Quested et al., 2003). Our annual green tea had between 50-55% mass loss (depending on snow regime), which is comparable to *Rubus chamaemorus*, but not to *Bistorta*, which is a dominant species at the Adventdalen snow fences. The annual mass loss for rooibos tea was around 25% for all snow regimes, and is comparable to decomposition of the dwarf shrub litter in Abisko.

In 2007, Cornelissen et al. used the same litter beds in Abisko as Quested et al. (2003) to compare a more mild Arctic decomposition study to a colder site in Latnjajaure (Cornelissen et al., 2007). This was a two-year litter decomposition study beginning in August, where forbs showed

85% mass loss in Abisko, and 60% mass loss in Latnja. Deciduous and evergreen shrubs were around 50% in Abisko and 35% in Latnja. Both the forbs and shrubs have higher decomposition values than our green and rooibos tea bags buried for one year, but this will be an interesting data set to compare to the remaining tea bags at our site that will be removed from the Adventdalen snow fences this June (future analysis for two years of decomposition).

Another snow fence study in Greenland assessed the effect of snow and of OTCs on winter, winter and spring, and winter, spring and summer mass loss of arctic plant litter (Blok et al., 2016). Blok et al. found significantly higher mass loss in both *Salix glauca* and *Cassiope tetragona* in the plots with increased snow depth compared to ambient. The values of decomposition for *Salix* (5-10%, depending on the period of decomposition and the snow treatment) and *Cassiope* (12-20%) are slightly lower than the values for our winter decomposition in rooibos (22%) and for annual decomposition (27%), but are quite comparable. The OTCs used in Blok's experiment caused a decrease in decomposition inside of OTCs, which is aligned with my findings for rooibos tea bags buried inside of OTCs during the summer period.

Although it is difficult to compare my high Arctic study site to lower Arctic litter decomposition experiments, these comparisons do imply that the decomposition of green tea is potentially higher than what would be exhibited by native arctic forb species in the high Arctic, while the rooibos tea bags might be a relatively accurate surrogate for arctic shrub decomposition.

6 Conclusions

In order to improve this study in the future, and to eliminate possible errors, soil samples should be taken and microbial analysis should be performed to identify any potential shifts in microbial community composition. In the springtime, tea bags should also be retrieved from the soil at the immediate completion of snowmelt for each snow regime, and temperature sensors should be secured 1cm under the soil surface. Soil samples could also be taken to facilitate a laboratory experiment for tea bag decomposition. Moisture, temperature, and microbial community composition could be more closely monitored in this type of lab experiment. However, the results from an *in situ* experiment offer valuable ecological insight, especially over the duration of many years.

Together, temperature, moisture, plant litter type and quality, and the microbial community play an important role in determining decomposition processes in soils around the world. This complexity makes it apparent that there are many knowledge gaps in the relationship between these factors, and leaves many questions yet to be answered. This study was conducted with limited time, and only shines light on a small portion of the complex issues of decomposition in the context of climate change. Originally, the experimental setup included three replicates per plot for annual decomposition rates. However, only one of these tea bags was removed after one year to observe winter decomposition, and the other was left in the soil to eventually record a two-year period of decomposition. These remaining tea bags may offer valuable insight into long-term decomposition with varying snow depths in the high Arctic.

The sampling error in harvesting tea bags regardless of the timing of snowmelt for each snow depth regime may have decreased the ecological significance of the summer and winter data sets. However, the consequent questions procured from the results of this experiment are important ones: How does the timing of snowmelt affect the processes of decomposition in Arctic soils? How have microbial communities in the various snow depth regimes at the Adventdalen snow fences changed over the past 13 years? Is this tea bag experiment an appropriate proxy for realistic decomposition rates of Arctic plant species? Although similarities between rooibos and green tea and Arctic species can be found, the use of tea bags for this experiment is only an estimation for what might potentially happen in the future. Additional studies of tea bags and decomposition under controlled experimentation would be necessary to understand any potential biases. In order to confidently understand this complex ecosystem, indigenous plant litter would be an asset to concluding these results. Decomposition must be studied at both an ecosystem scale and at a microbial scale to truly understand the influence a dynamic development of climate change could have on Arctic soils, and the life they support above and below the soil surface.

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Figure A1. Annual daily minimum temperatures outside OTCs for all snow regimes.



Figure A2. Annual daily maximum temperatures outside OTCs for all snow regimes.



Figure A3. Anual below zero temperatures outside OTCs for all snow regimes.



Figure A4. Summer mean, minimum, and maximum hourly temperatures for a 24hr period (doy=200) outside OTCs for all snow regimes.



Figure A5. Summer minimum and maximum hourly temperatures for a 24hr period (doy=200) outside OTCs for all snow regimes.



Figure A6. Soil moisture measurements in September, (a) outside OTCs (29 observations) and (b) inside OTCs (29 observations) for all snow regimes showing estimated means and their 95% confidence intervals.



Figure A7. Soil moisture measurements for all snow regimes taken in (a) June, inside and outside of OTCs (59 observations) and (b) in September outside OTCs (29 observations) showing estimated means and their 95% confidence intervals.



Figure A8. Soil moisture measurements for all snow regimes taken in (a) June, inside and outside of OTCs (59 observations) and (b) in September inside OTCs (29 observations) showing estimated means and their 95% confidence intervals.