Evaluating the Tea Bag Index approach for different management practices in agroecosystems using long-term field experiments in Austria and Sweden

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Abstract

Litter decomposition is an important factor affecting local and global C cycles. It is known that decomposition through soil microbial activity in ecosystems is mainly influenced by soil type and climatic conditions. However, for agroecosystems, there remains a need for a better understanding how management practices influence litter decomposition. This study examined the effect of different management practices on decomposition at 29 sites with long-term (mean duration of 38 years) field experiments (LTEs) using the Tea Bag Index (TBI) protocol with standard litter (Rooibos and Green tea) developed by Keuskamp et al. (2013). The objective was to determine if the TBI decomposition rate (*k*) and stabilization factor (*S*) are sensitive enough to detect differences in litter decomposition between management practices, and how they interact with edaphic factors, crop type and local climatic conditions. Tea bags were buried and collected after ~90 days in 16 Austrian and 13 Swedish sites. The treatments at Austrian LTEs focused on mineral and organic fertilizer application, tillage systems and crop residues management, whereas those in Sweden addressed cropping systems, mineral fertilizer application and tillage systems. The results showed that in Austria, incorporation of crop residues and high N fertilizer application increased *k*. Minimum tillage had significantly higher *k* compared to reduced and conventional tillage. In Sweden, fertilized plots showed higher *S* than non-fertilized plots and high N fertilizer had the highest *k*. Growing spring cereal lead to higher *k* than forage crops. Random Forest regressions for Austria and Sweden jointly showed that *k* and *S* were mainly governed by climatic conditions, which explained more than 70% of their variation. However, under similar climatic conditions, management practices strongly influenced decomposition dynamics. Thus, the TBI approach may

be suitable to apply in a more large-scale network on LTEs for evaluating decomposition dynamics in agroecosystems more precisely.

Introduction

Soil organic carbon (SOC) is one of the most used indicators for soil quality, since its dynamics is involved in regulating ecosystem functionality through its influences on physical, biological and chemical soil properties, which are critical for nutrient cycling and soil fertility (Davidson et al. 2006; Janzen 2015). Management practices, such as fertilizer application, use of catch- and cover crops, organic amendments, length of bare fallow periods, permanent surface protection with perennial crops, tillage practices and aboveground crop residue management, are impacting SOC balances for agroecosystems (Kätterer and Bolinder, 2022; Sandén et al., 2018; Paustian et al., 2016).

Agricultural soils play a crucial role in the global carbon (C) cycle due to their C sink capacity (Paustian et al., 2016; Lal, 2004). In this context, improved management practices that maintain or increase SOC stocks are considered essential in national greenhouse gas reporting systems (IPCC, 2006), as well as in other international incentives such as the 4 per mille initiative (Minasny et al., 2017). The SOC balance is dynamic and determined by the difference between annual C inputs to soil, and the annual C outputs through the decay of existing soil organic matter and resulting from microbial activity, which is the main contributor to stable SOC (Tiefenbacher et al., 2021; Bolinder et al., 2007). Management practices have a great impact on these two factors by affecting either the amount of C inputs or outputs through decomposition, or both factors simultaneously.

Litter decomposition is a complex biogeochemical process controlled by several biotic and abiotic factors, where the biological activity of decomposers varies with soil properties and is driven largely by climatic conditions (Daebeler et al., 2022; Bradford et al., 2016; Cleveland et al., 2014; Gholz et al., 2000). Decomposition and SOC stabilization are long processes, therefore long-term field experiments (LTEs) are among the most useful resources for quantifying the impact of management practices on litter decomposition, SOC changes, and soil functioning (Sandén et al., 2018; Kätterer et al., 2012; Bergkvist and Öborn, 2011). Experiments determining litter mass loss over time *in situ* are also important for understanding SOC dynamics, nutrient cycling and colonization by soil biota under field conditions. The traditional method that has been used in ecology for more than 50 years consists of litterbag studies, burying known quantities of various organic materials into the soil, and retrieving them successively at different intervals (Kampichler and Bruckner, 2009; Burgess et al., 2002; Bocock and Gilbert, 1957). These studies are not always comparable because they are subject to variations in e.g., litter type, mesh-size, sample preparation and analytical methods, and the placement of litterbags may alter the microclimate for decomposers (Kampichler and Bruckner, 2009).

Keuskamp et al. (2013) developed therefore a low-cost and time-efficient methodology called Tea Bag Index (TBI), characterizing the decomposition process with commercially available tea bags, where green tea is representing labile organic material and rooibos tea as a surrogate for recalcitrant litter. A decomposition rate (*k*) and a stabilization factor (*S*) are obtained accordingly with their chemical composition and the respective weight lost at a single point in time after an incubation period of ca 90-days in the soil. The TBI approach is particularly useful for assessing geographical differences in decomposition dynamics because results are directly comparable across sites, varying only with local edaphic and seasonal environmental conditions (Keuskamp et al., 2013).

In several studies, the TBI has been used as an indicator for biological (Sandén et al., 2021; Costantini et al., 2018) and microbial activity (Daebeler et al., 2022; Treharne et al., 2019; Tóth et al., 2017), ecosystem functioning, nutrient cycling (Zaller et al., 2016), and soil quality (Tresch et al., 2018; Buchholz et al., 2017). Generally, this follows the concepts of soil quality as reviewed by Bünemann et al. (2018), where TBI would primarily be a biological soil quality indicator, although the data apparently were not normalized for the temperature effect. According to the TBI community, data collected from networks of researchers and citizen scientists for constructing a global TBI map (www.teatime4science.org) shows that most studies have been using the TBI approach for different forest and grassland ecosystems or urban soils, agricultural fields represent less than 15% of data. Indeed, there are only a few published studies (Daebeler et al., 2022; Dossou-Yovo et al., 2022; Struijk et al., 2022; Fu et al., 2021; Sandén et al., 2020; Barel et al., 2019; Poeplau et al., 2018; Sievers and Cook, 2018) that have been using the TBI approach for evaluating agroecosystems, and it is not clear if this method is sensitive enough to detect differences between management practices.

This study used the TBI approach for investigating the effect of management practices on the decomposition rate (k) and stabilization factor (S) at several LTEs in Austria and Sweden, with different soil characteristics, and climatic conditions, and subjected to various management/treatments.

To the best of our knowledge, this is the first analysis using the TBI approach for such a large number of LTEs and different treatments for agroecosystems. The treatments covered management practices such as organic amendments, crop rotations, aboveground crop residue handling, mineral fertilizer application, and tillage. Our objectives were: (i) to evaluate if the TBI *k* and *S* parameters are sensible enough to distinguish between different management practices; (ii) to quantify the effect of management practices on *k* and *S*; and (iii) to identify the most important local climate and/or soil properties affecting litter decomposition in Austria and Sweden.

Materials and Methods

Study sites

Austria

We used sixteen Austrian (AT) sites, by selecting contrasting treatments from three different categories of LTEs where the management practices had been in place for 11 to 63 years (see details in Table 1). The TBI measurements were made in 2014, 2015 and 2016. Measurements sometimes took place in more than one year at the same LTE (e.g., MUBIL), and the sites were abbreviated as AT1 to AT16. Six experiment categories involved carbon balance practices (CB) focusing on organic matter inputs such as compost and crop residues, eight sites were studying soil fertility (SF) in terms of differences in mineral N and P fertilization, and two sites examined tillage systems (TS). The sites are located in several agricultural areas across the country (Fig. 1), with diverse soil textures (Table S1) and variable crop types (Table 1) and climatic characteristics (Table 3) during the years of TBI measurements. More details for some of the sites are available in specific publications: AT3 (Spiegel et al., 2018; Lehtinen et al., 2017; Tatzber et al., 2015; Aichberger and Söllinger, 2009), AT4 to AT6 (Spiegel et al., 2018; Lehtinen et al., 2014), AT15 and AT16 (Tatzber et al., 2015; Spiegel et al., 2007). In addition, Sandén et al. (2018) puts the Austrian LTEs in the context of other European LTEs.

The purpose of the Austrian C balance LTEs was threefold: i) to assess the sustainability of stockless vs. livestock-keeping organic farming management on soil and crop traits (AT1, AT2), ii) to investigate the effects of compost amendments on soil and crops (AT3), and iii) to compare crop residue incorporation and removal (AT4-AT6). Compost amendment LTE and crop residue incorporation LTEs also included mineral fertilizer application, whereas AT1 and AT2 only focused on different organic fertilizer treatments.

The eight soil fertility LTEs (AT7-AT14) were all focusing on the effect of mineral fertilizer on soil and crop properties. In most cases, treatments studied different amounts of mineral nitrogen fertilizer, whereas AT9 and AT12 also investigated the effect of different amounts of K fertilizer application. Nitrogen fertilizer was applied in four stages and potassium in three stages, according to Austrian guidelines for fertilizer (BMLFUW, 2017).

For the tillage system experiments, conventional tillage (CT) was compared to reduced tillage (RT) and minimum tillage (MT). Regular mouldboard ploughing to 25–30 cm soil depth was applied in CT treatment, whereas cultivator in autumn to a depth of 15–20 cm was used in RT treatment and a rotary driller that loosened the soil to a depth of 5–8 cm was used in MT treatment. The soil was turned over only in the CT treatment, where inversion tillage was incorporating the crop residues. Fertilizer application was crop specific according to the Austrian guidelines for fertilizing (BMLFUW, 2017).

Sweden

We used thirteen Swedish (SE) sites by selecting contrasting treatments from three different categories of LTEs, where the management practices had been in place for 11 to 59 years (see details in Table 2). TBI measurements at these sites were made only in one year (2016), and were abbreviated as SE1 to SE13. Six sites involving combined management practices (CMP), four studying the effect of rotations (ROT), and three sites with tillage systems (TS). The sites are located in several agricultural areas across the country (Fig. 1), with diverse soil textures and variable crop types (Table 2) and climatic characteristics (Table 3) during the year of TBI measurements. Bergkvist and Öborn (2011) give a general description of all these LTEs. Only a brief description of treatments are provided below, more details on the sites with combined management practices is given by Carlgren and Mattson (2001), and for tillage systems by Arvidsson et al. (2014), while Poeplau et al. (2015) provide some more insight on the rotation experiments.

The initial purpose of the six LTEs with combined management practices (SE1-SE6) was to compare a change from the traditional mixed farm production system including crops and livestock into a pure cash crop system, by studying their effects on the sustainability of crop production and soil properties (entitled *soil fertility experiments*). The dairy production treatments contain exclusively perennial grass-clover leys and receive one farmyard manure (FYM) application per rotation. The cash crop treatments consist of annual crops (i.e., oilseed is replacing leys in the rotation) without manure applications (0 FYM) only receiving mineral fertilizers application (NPK). PK applications in all the treatments we selected were aimed at achieving rapid build-up of the soil PK status, i.e., the amount applied was first replacing that exported in harvested products (i.e., maintenance principle), to which an extra amount was added (corresponding to the max treatment). The N-rates in all NPK treatments were also corresponding to max application rate, and were adapted depending on crop type, where spring cereals, oilseeds, and leys received 125 kg, while sugar beet received 210 kg N ha^{-1} yr^{-1}. We were also using the control plots receiving no NPK (0 NPK). As a third factor, aboveground crop residue removal takes place in all FYM treatments, simulating use of harvest residues for fodder or bedding material that are recycled as manure. The southern sites have 4-year rotations and those in central Sweden have 6-year rotations. The north site (SE5) is slightly different from the others, consisting of a 7-year rotation and is studying only the livestock-based production system.

For rotation experiments purposes, we were comparing extreme treatments representing two rotations from four LTEs (SE7-SE10) with the main objective to study changes in SOC (named *humus balance experiments*), i.e., a continuous spring cereal (SC) system and a ley-dominated rotations (L). The straw was removed from the plots every year in the SC treatments, and L consisted of a grass-clover mixture re-established every fourth year. Both rotations were receiving P and K accordingly with the maintenance principle, and SC and L were receiving 120 and 150 kg N ha^{-1} yr^{-1}, respectively.

In the three tillage experiments (SE11-SE13), the conventional tillage (CT) and direct seeding (DS) treatments were the same for all sites, consisting of inversion plowing to a depth of 20-23 cm and by using a disc seed drill, respectively. The shallow (5-7 cm) and deep (~12 cm) reduced tillage treatments (SRT and DRT, respectively), consisted of primary tillage operations made in the autumn and most commonly with a chisel plough. The main crops in all the tillage system experiments were winter and spring cereals (occasionally oilseed), fertilized accordingly with local recommendations and with the aboveground residues chopped and left in the field.

TBI method and sampling design

The TBI method was used according to the protocol established by Keuskamp et al. (2013) to determine litter decomposition using two types of commercial tetrahedron-shaped tea bags by Lipton Unilever (Green tea and Rooibos tea). The green tea (*Camellia sinensis*; EAN:

8722700055525) has high cellulose content, higher soluble fraction, and lower C:N ratio; while rooibos tea (*Aspalathus linearis*; EAN: 8722700188438) has high lignin content, lower soluble fraction, and higher C:N ratio, which is expected to slow down decomposition (Keuskamp et al., 2013). The synthetic tea bag material has a mesh size opening of 0.25 mm allowing access to microorganisms, very fine roots and root hairs.

The initial mass of the tea bag contents was determined on 20 randomly selected bags for each tea type from different boxes, oven-dried at 70^{o}C for 48 hours and weighed separately; the mean dry mass for green tea was 1.717 ± 0.048 g and that for rooibos tea was 1.835 ± 0.027 g. For both countries, close to seeding of annual crops, from end of April to mid-June depending on location, each tea bag was properly identified and buried in the soil at 8 cm depth.

For Austria, four bags of each tea were used and placed side by side at a distance of 2 to 3 cm. Each tea bag was properly identified and buried in the soil at 8 cm depth. The TBI incubation period from placement to last retrieval averaged 80±13 days (Table 3) due to logistic issues for collection. After collecting, the tea bags were oven-dried at 70^{o}C for 48 hours after removal of adhered soil particles according to the standardized protocol by Keuskamp et al. (2013). After drying, the tea bags were opened and the tea content was weighted. The ash content was not determined.

The same TBI protocol was used for the Swedish sites. As in Austria, four bags of each tea were used per experimental unit, placed side-by-side at a distance of 2 to 3 cm. Each tea bag was properly identified and buried in the soil at 8 cm depth. The mean TBI incubation period from placement to last retrieval date averaged 91±1 day (Table 3). In addition to the removal of adhering soil particles, the ash content was determined (i.e., both for green and rooibos tea on mixed samples of the four replicates) in a muffle oven at 550^{o}C for 16 hours. The rational for measuring the ash content was that three of the Swedish sites had a high clay content (Table S1), where the complete removal of adhering soil particles may be more difficult. However, the ash content was on average quite low, representing 15±6% and 10±4% for the Green and Rooibos tea bags, respectively (data not shown).

After measuring the remaining dry matter, the decomposition rate (*k*) and stabilization factor (*S*) for both countries were calculated according to the TBI presented by Keuskamp et al. (2013). This standardized method that is using single measurements after an incubation period in the soil of 90-days have received some criticism. For instance, Mori (2022) and Mori et al. (2023) showed that this incubation period is not always long enough for the mass loss of green tea to reach a plateau, and further suggested that time-series mass loss data of rooibos tea is also required to respect the underlying assumptions of the TBI method. Time-series (15, 30, 60 and 90-days) of green and rooibos tea were available for all the Swedish sites but only at one Austrian site (16, 26, 62 and 91-days at AT16). The incubation period was consistently always 90-days for the Swedish sites, and only shorter than that (i.e., about 60-days incubation period) for a few of the Austrian sites (Table 3). To have as uniform comparisons as possible between the two datasets, we only used the last measurement for both countries for calculating *k* and *S*. The purpose of using the time-series for testing the underlying TBI assumptions was beyond the scope of this paper.

The daily climate data for Austria were retrieved from the Central Institution for Meteorology and Geodynamics (ZAMG). For Sweden, the daily climate data were gathered through official data from the most nearby LantMet climate stations, and from the Swedish Meteorological and Hydrological Institute (SMHI). The climate variables used in this study were air temperature, precipitation, solar radiation, wind speed and air humidity (Table 3).

For Austrian soils, pH was measured electrochemically (pH/mV Pocket Meter pH 340i, WTW, Weilheim, Germany) in 0.01 M $CaCl_2$ at a soil-to-solution ratio of 1:5 (ÖNORM L1083). Total soil organic C (TOC) concentrations were analyzed by dry combustion in a LECO RC-612 TruMac CN (LECO Corp., St. Joseph, MI, United States) at 650°C (ÖNORM L1080). Total N (Ntot) was determined according to ÖNORM L1095 with elemental analysis using a CNS (carbon, nitrogen, sulfur) 2000 SGA-410–06 at 1250°C. Texture was determined according to ÖNORM L1061-1 and L1062-2. For Sweden the data were gathered from recent archived analysis protocols (pH was measured in water). Clay content, C content, C:N ratio, and pH measured from each site in both countries are shown in Table S1 (Supplementary material).

Data analysis

Analysis of variance (ANOVA) for each experiment category (i.e., CB, SF, TS, CMP and ROT) was performed to analyze the effects of the treatments and the differences between sites on *k* and *S* separately for both countries. When the treatments were identical within the same experiment category, sites were used as a random effect with a mixed ANOVA to test the average treatment effect, mean values were used as replicates to test the differences between sites. The Tukey's test ($p < 0.05$) was used for comparing the same treatments and the same sites using R software version 4.2.2. We have treated the data from different years at the same sites in Austria as independent observations, in the sense they are not a time series of measurement. Interactions between site and treatment were considered.

We calculated a climate-dependent soil biological activity parameter (Re_{clim}), by using mean daily air temperature, total precipitation, and potential evapotranspiration (PET) data in pedotransfer, soil water balance and biological activity functions. Compared to raw climatic data alone, this parameter is integrating the effect of climate, soil and crop properties. It is calculated as the product of a soil temperature (Re_{temp}) and relative water content (Re_{wat}) factor with a daily time step (i.e., $Re_{clim} = Re_{temp} \times Re_{wat}$), which is thereafter averaged to give an estimate of soil biological activity for a given time period. These two factors are derived from soil temperature and soil moisture response functions expressing the activity of decomposers and their relative effect on the decay rates of organic materials in the arable layer of agricultural soils. Briefly, the Re_{temp} is calculated from air temperature and leaf area index using an empirical model (Kätterer and Andrén, 2009), while Re_{wat} is calculated using pedotransfer functions for simulating the soil water balance and a function for estimating PET. In addition to air temperature and leaf area index, calculations of Re_{wat} also involve the use of daily climatic data for precipitation, wind speed, air humidity, and solar radiation, as well crop types and yields, soil texture and SOC content. For details see Bolinder et al., 2008, and Fortin et al., 2011) and information in the following R package used for the calculations: https://github.com/ilmenichetti/reclim (Please refer to the package documentation). The Re_{clim} concept can be used for quantifying regional differences in soil biological activity alone (Bolinder et al., 2013; Andrén et al. 2007) or integrated as a parameter in the ICBM SOC model. In the latter case, it is adjusting the decomposition rates of both C inputs to soil from crop residues (e.g., straw) and that of the more stable SOC (Andrén and Kätterer, 1997; Andrén et al., 2004). In this study, we used the concept of Re_{clim} and the Re_{temp} and Re_{wat} factors to test if the product of soil-temperature and relative water content better explained the variation in k and S than the two latter alone, and to determine if they also better explained this variation compared to using only raw climatic data.

We calculated simple correlation between variables mean annual temperature (MAT), mean temperature during the incubation period (MAT_{TBI}), total annual precipitation (TAP), total precipitation during incubation period (TP_{TBI}), potential evapotranspiration (PET), potential evapotranspiration during incubation period ($\mathrm{PET}_{\mathrm{TBI}}$), aridity index (AI), aridity index during incubation period ($\mathrm{AI}_{\mathrm{TBI}}$), temperature and precipitation factor (TxP), Re_{clim}, Re_{wat}, Re_{temp}, pH, SOC, clay and C:N ratio, using Pearson correlation. For more accurate results, we applied random forest (RF) regression in order to rank the importance of variables for *k* and *S*, using the random forest R package (Liaw and Wiener, 2002). The RF is a machine learning technique based on decision trees that predicts a certain variable from a set of other variables through a series of binary splits of the data, where the variables are either continuous or categorical. For example, in the case of a continuous variable it consists of all the data points above or below a certain threshold, for a categorical variable it consists of all the data points belonging or not to a specific class. All these subsequent splits constitute a decision tree. A random forest is a set of decision trees and it is therefore an ensemble technique. This allowed us to utilize treatment and crop variables (including N fertilizer) without having to convert them into a ranking. Another useful asset of an RF regression is that it evaluates the importance of each variable in defining the predicted variable. There are various possible measurements to do that, but they are all based on measuring the effectiveness of each subsequent split in each node of a decision tree in sorting out the information. In our study, we used a measurement called node purity based on the Gini index, which expresses the probability of one split of the data (i.e., one node of the tree) defining the predicted variable. The total node purity of a certain variable in a tree is the sum of all the node purity measurements for each node considering that particular variable, and the higher it is the more that variable is important.

We used the following models to predict the two TBI parameters *k* and *S*:

$$k_n\ f(TN, SOC, CN, cl, cr, tr, pH, N, PET, PET_{TBI}, TAP, TP_{TBI}, MAT, MAT_{TBI}, AI, AI_{TBI}, Re_{clim}, Re_{wat}, Re_{temp})$$

$$S_n\ f\left(TN, SOC, CN, cl, cr, tr, pH, N, PET, PET_{TBI}, TAP, TP_{TBI}, MAT, MAT_{TBI}, AI, AI_{TBI}, Re_{clim}, Re_{wat}, Re_{temp}\right)$$

where soil variables (continuous) were TN (total N g kg^{-1}), SOC (g kg^{-1}), CN (C:N ratio), cl (clay content g 100 g^{-1}) and pH. Categorical variables were N (N fertilizer application factor with 4 levels), cr (crop factor, e.g., barley, ley (establishment), ley (production), oat, spring oilseeds, sugar beet, maize and winter wheat) and tr (a treatment factor with 30 levels). The climatic variables (PET_{TBI}, PET, TP_{TBI}, TAP, MAT_{TBI}, MAT, AI_{TBI} and AI) are as defined in Table 3. The climate response variables Re_{clim}, Re_{wat}, Re_{temp} are as described above.

Since many of variables in our model are likely to be correlated and carry similar information, we applied the recursive feature elimination algorithm implemented in the caret R package by Kuhn et al. (2016), which assess in subsequent iterations the optimal set of predicting variables (features) to be utilized by the RF model. The procedure starts by fitting a RF model with all variables, ranking them by importance, and discarding the least important. The algorithm then iterates. The optimal number and set of features are then defined by a fitness metric (in our case the model R^2), selecting the set with the best model fitness. The selected models were used to compute the variables' relative importance.

In order to better understand the similarities between the sites, we run a principal component analysis (PCA) in the space composed by the variables pH, SOC, TN, C:N, clay, TAP, MAT, PET, AI and TxP. The analysis was run with the R command prcomp (Venables and Ripley, 2002), from the base R installation (“Stats” package). Variables were all standardized by rescaling them to zero mean and unit variance.

Results

Effect of management practices

Austria

Both the TBI parameters *k* and *S* varied between treatments and sites in Austria, and even between years at the same site within the C balance category (Fig. 2 and Table S2). In general, all treatments in AT1 with a lucerne crop under wetter conditions (2014) presented higher *k* and lower *S* than in AT2 with a wheat crop under dryer conditions (2015), and the treatment with municipal compost and green manure (FW) had the highest *S* in 2015. The AT3 site did not present significant differences between the treatments. Crop residue incorporation treatment (CRI) had higher *k* than the crop residue removal (CRR) at the AT4 and AT6 sites, and AT6 presented a higher *k* than at AT4. Comparing years for the same experiment site and type with different crops, AT5 (2015; with wheat) had higher *S* than AT6 (2016; with maize).

For the soil fertility experiment category (Table 1), AT12 had the highest *k* and AT13 had the highest *S*. Sites receiving NPK fertilizer application (AT7, AT8 and AT9) had higher *k* and *S* even at different N doses than at the AT12, AT13 and AT14 sites receiving only N (i.e., without P and K). Stabilization was significantly higher in AT9 than in AT7. For potassium trials, AT11 presented significantly higher *k* and *S* than AT10. Regarding sites receiving only N addition (AT12, AT13, and AT14), maximum doses (180 kg N) presented the highest *k* (0.0095), and no N addition had the lowest *k* (0.0066) (Fig. 2 and Table S2).

Regarding the tillage system experiment category (AT15 and AT16 - Table 1), *k* was significantly higher in shallow reduced tillage (SRT) and *S* was higher in deep reduced tillage (DRT) in 2015 (AT15), but no significant differences between treatments were found in 2016 (AT16). Site AT16 had significantly higher *S* than AT15 (Fig. 2. and Table S2).

Sweden

At the Swedish sites (Table 2), the TBI measurements for the combined management practices experiment category showed that both *k* and *S* were significantly higher for the FYM/NPK treatments maximum amount of farmyard manure and maximum doses of NPK (FYM/NPK) (Fig. 3, Table S3) compared with the control treatments (0 FYM/0 NPK). Comparing sites, SE5 and SE6 had the highest *k* and SE4 had the lowest *k*, while SE3 presented the highest *S* followed by SE6, SE5 and SE4, the lowest *S* was for SE1 and SE2 (Table S3).

Regarding the rotation experiments, the continuous spring cereal rotation presented higher *k* than for ley, but there was no significant difference in *S*. Comparing sites, SE9 presented higher *k* than SE7, SE8 and SE10, whereas SE7 and SE8 had the highest *S*.

For the tillage system experiment category, conventional tillage (CT) had the lowest *k* and *S*, while deep reduced tillage (DRT) had the highest *k* and *S*. The highest *S* was observed for the SE12 followed by SE11 and SE13. Sites did not show significant differences for *k*.

Comparing tillage system experiments in Austria and Sweden (2016) for sites (AT16, SE11, SE12 and SE13) for the treatments that were the same in both countries (CT, SRT and DRT), deep reduced tillage had the highest *k* and shallow reduced tillage had the highest *S*. The Austrian site presented the lowest *k* and the highest *S* compared to the Swedish sites, which did not present significant differences among them (Table S3).

The field application of the TBI found a clear discrimination of both *k* and *S* values between the two countries. The both mean *k* and *S* values were higher in Sweden (Table 4, Fig. 4). In general, the variation in *k* and *S* values were lower in Austria. Indeed, mean *k* by site in Austria varied between 0.0058and 0.0128 and mean *S* varied between 0.113 and 0.442 (Table S2). Whereas mean *k* by site in Sweden was between 0.0084 and 0.0301, and mean *S* was between 0.118 and 0.361 (Table S3). All values for *k* and *S* were within the range of the previous global TBI investigation (0.005-0.04 for *k*; and 0.05-0.55 for *S*) by Sandén et al. (2020).

Influence of climate and soil properties

Using the combined dataset for Austria and Sweden, resulted in significant negative correlation between *k* and MAT, TAP, PET, TxP factor, Re_{clim} and Re_{temp}, and significant positive correlation with the C:N ratio (Table S4). Stabilization factors in Austria and Sweden combined correlated negatively with MAT_{TBI} period, Re_{clim} and Re_{temp}.

The variable selection procedure with the random forest models (Fig. 5) identified fewer variables explaining *k* and *S* values for the combined dataset. The variables explaining *k* is low, but the overall predicting power of the model decreased substantially.

According to the optimized random forest model, most of the variance in *k* was accounted for by climatic variables (Fig. 5), with Re_{temp} ranking the highest, followed by AI and MAT. It was also showing that treatment, C:N ratio and SOC were important factors as well. The *S* was also influenced by climate-related variables, withRe_{temp} leading the ranking, and it was followed by pH and more agronomic variables such as treatment, crop and N fertilizer application.

The principal component analysis (Fig. 6) revealed that the data are well divided in two groups, representing the two countries, which allow us to say that indeed edapho-climatic characteristics are different between Austria and Sweden. PC1 showed that points in Austria are positively related to climatic characteristics, such as MAT, TAP and PET. On the other hand, points in Sweden are positively influenced by high clay content and high C:N ratio. High C:N enhancement is related to low TAP and TxP. High clay contents are related to low MAT and PET, which can be due to an historical influence on soil weathering. PC2 is dominated by SOC and TN contents, mainly at two points in Austria (AT7 and AT9: two sites testing soil fertility with NPK and relatively low TAP and TP_{TBI} compared to other sites). Austria presented more divergent data, having more heterogeneous sites than in Sweden.

Discussion

Influence of climate and soil properties

In previous studies using the TBI approach, it was shown that climate played a significant role on decomposition in a temperate biome (Djukic et al., 2018), but when comparing several different biomes, climatic conditions were of relatively low importance (Fanin et al., 2020). In Boreal soils, Althuizen et al. (2018) found that increased temperatures enhanced k, whereas increased precipitation decreased k across years. Many studies have showed a positive correlation between precipitation and decomposition rates (Pimentel et al., 2019; García Palacios et al., 2016), however precipitation did not show correlation with k according to the random forest analysis. On the other hand, Re_{wat} showed great importance (Fig. 5). It is because this variable includes nonlinearities due to its shape according to which decomposition increases with soil moisture and then decreases at high soil water content due to oxygen limitation of microorganisms (Moyano et al., 2013).

In general, higher k values were observed when the aridity index was lower, and it was identified by the random forest regression model being an important variable affecting the rate of decomposition (Fig. 5). This may be related to the observations that in more arid and warmer sites, the biological processes driving SOC dynamics are impaired which may result in decreasing soil C and N stocks (Kerr and Ochsner, 2020; Ontl and Schulte, 2012; Jiao et al., 2016; Reynolds et al., 2007).

For practical reasons the teabags in our study were buried in the soil during the growing season, corresponding to a period when the soil biological activity is highest (Bolinder et al., 2013). When burying the teabags during the growing season, the difference in climatic variables between sites are attenuated, in particular with respect to air temperature. For example, the MAT at the Swedish most northerly (SE5 and SE9) and southerly (SE2) sites are 4.1 and 9.0 ^{o}C, respectively, whereas the corresponding mean air temperatures during the incubation period (MAT_{TBI}) were 14.3 and 16.2 ^{o}C, respectively.

The TBI *S* parameter was also dependent on climate. Indeed, the random forest model identified climatic parameters as the main factors affecting *S* in both Austria and Sweden during all evaluated periods. In particular, Re_{clim} and Re_{temp} often showed significant negative correlations, which implies a negative impact of air temperature on *S*. However, raw climatic variables, such as precipitation and temperature were only weakly correlated with *S*. This is probably due to nonlinear processes, which are considered in the ICBM climate-dependent soil biological activity calculations such as Re_{wat} (as discussed above). Furthermore, since litter decomposition dynamics is influenced by multiple factors that interact and change over time (Bradford et al., 2016), the relationships are not always linear. Random forest models that we fitted to the data are more efficient capturing such combinations and interactions of factors, and can detect relationships that would not be detectable by linear approaches.

Effect of management practices

Our results revealed that a large number of different management practices significantly affected both the decomposition rate *k* and stabilization factor *S* according to the TBI approach used in several LTEs in Austria and Sweden (Fig. 2, 3 and 5). This is in contrast to the studies by Djukic et al. (2018) as well as Saint-Laurent and Arsenault-Boucher (2020), who did not find any significant effect of land use and management practices on early-stage litter decomposition in a temperate biome.

In the C balance trials in Austria, soils receiving green manure + municipal compost had higher *S* than soil receiving biogas slurry at the second year (Table S2). This is in agreement with studies indicating that compost can improve SOC stabilization over time (Mekki et al., 2019; Eshetu et al., 2013; Ceccanti et al., 2007). The higher *k* in the crop residues incorporated treatments (AT4 and AT6; Table S2) can be attributed to the fact that incorporation of crop residues into the soil can increase the decomposition rate by stimulating microbial activity. During the early stages of decomposition, soluble C is rapidly utilized by soil biota (Werth and Kuzyakov, 2010). The higher *k* and lower *S* at AT6 compared to AT4 were likely due to the loamy texture, lower potential evapotranspiration resulting in a lower aridity index at the AT4 site (Table 3 and Table S1), corroborated by the PCA analysis (Fig. 6).

There were no significant differences in *k* and *S* found among treatments in the soil fertility trials in Austria with NPK addition. However, there was a trend towards a higher *S* at AT9 compared to AT7, likely related to the higher SOC content in AT9, since the climatic conditions and soil texture were quite similar for both areas, which suggests that higher SOC content may have increased *S*. Site AT11 had higher *k* and *S* than AT10. Possible explanations for this trend are that AT10 had lower clay content, lower precipitation resulting in higher PET (corroborated by the PCA analysis) and AI, contributing to a lower soil moisture content and thereby lower decomposition and stabilization.

Nitrogen supply may favor microbial activity and thereby litter decomposition (Raiesi, 2004). This was reflected in the treatments where only N was added, where the high dose of 180 kg N ha^{-1} (AT12, AT13 and AT14) induced a significantly higher *k* (Table S2), compared to the treatments with no N addition, which also had the lowest *k*. Furthermore, the significant difference between sites, in which AT12 had the highest *k* could at least partly be explained by a higher SOC and higher pH at this site (corroborated by the PCA analysis).

In the tillage system experiment at Fuchsenbigl in Austria in 2015 (AT15, with maize) and 2016 (AT16; with wheat), the shallow reduced tillage showed significantly higher *k* than deep reduced tillage and conventional tillage, only in 2015, indicating that shallow soil tillage stimulated decomposition that particular year. This was likely due to climatic conditions, since 2015 was slightly drier and warmer, furthermore maize straw has lower C:N ratio than wheat straw and tends to decompose faster. Some studies showed faster decomposition under conventional tillage than under reduced tillage practices (e.g., Lupwayi et al., 2004). However, Kainiemi et al. (2015) found a decrease in soil respiration in conventional tillage compared to shallow tillage in temperate regions, which directly implies a lower decomposition (and lower *k*). These differences between tillage treatments are attributable to indirect effects on soil moisture and temperature profiles. We attribute the significantly higher *S* in 2016 to the fact that this year was moister and less warm, compared to 2015, resulting in a lower aridity index during the TBI incubation period (AI_{TBI}).

In the Swedish combined management practices trials, treatments receiving organic and mineral fertilizer application had higher *k* and *S* (FYM/NPK; Table S3), likely due to the increase in microbial diversity and activity favored by nutrient and C supply (Stark et al., 2007). Sites SE5 and SE6 presented the highest *k*: SE5 had low evapotranspiration and ardity index, resulting in more moisture; SE6 had also high *S*, due to high clay content and low PET (corroborated by the PCA analysis). Site SE3 had high *S*, which could be related to a higher C:N ratio, as suggested in another TBI experiment by Althuizen et al. (2018), a study showing that C:N ratio is positively correlated to *S*. The SE1 and SE2 sites had lower *S* than SE3 despite similar climatic conditions, which probably was related to the type of crops growing in these treatments (i.e., sugar beet in the SE1 and SE2 and grass/clover ley in SE3), since these two crop types have different effects on soil temperature and moisture.

In the Swedish rotation system trials, spring cereal (SC) had higher *k* than ley (Table S3). Site SE9 had higher *k* and lower *S*, in which the low stabilization may be caused by low clay content, low pH, and high solar radiation, leading to low SOC (corroborated the PCA analysis). The highest *S* were found in sites SE7 and SE8, in which the former presented high clay and SOC content, and SE8 had high precipitation and low PET.

For the tillage system treatments in Sweden, similar to the Austrian sites, the conventional tillage presented the lowest *k*, and also lowest *S*. Even when comparing tillage systems in Sweden and Austria jointly (Table S3) we could notice that conventional tillage also presented the lowest *k*, while deep reduced tillage the highest.

The decomposition rate *k* was mostly affected by climatic conditions, which was already expected, as temperature and aridity or moisture, however agronomic and edaphic factors proved to be of great relevance for *k*, as soil management choice (treatment), soil C:N ratio, SOC, crop type, pH, clay content, soil nitrogen and others. The stabilization factor *S* expresses the degree by which the labile fraction of the plant material is decomposed. We observed that the influence of pH was the most important edaphic factor, and the agronomic factors as the soil management treatment, crop type, and N fertilizer application were also identified as good predictors. A study conducted by Fu et al. (2021) suggested that pH, nutrient availability and soil compaction were the main reasons contributing to the differences in litter decomposition. The net effect of pH is not clear since it modifies both SOC decay kinetics and productivity simultaneously (Paradelo et al., 2015), with a maximum effect at around neutral pH (Liao et al., 2016). Nevertheless, the impact of pH on litter decomposition using the TBI approach seems clear in our study.

Conclusion

Our results show that both the TBI *k* and *S* parameters were sensitive to management practices in agroecosystems in Austria and Sweden. We were observing significant differences for some of the treatments in all categories of LTEs. Notably, for the effect of crop residue incorporation, organic amendments and N fertilizer application, crop types and tillage systems. In the Austrian LTEs, application of green manure + municipal compost showed a higher *S* compared to the application of other organic amendments. Incorporation of crop residues and high N fertilizer application also increased *k*. In the Swedish LTEs, it was shown that combined management practices with both farmyard manure and mineral NPK resulted in higher *k* and *S* compared to no manure and no NPK applications, whereas growing spring cereals instead of leys increased *k* but did not change *S*. For both countries, tillage systems with deep reduced tillage practices presented higher *k*, and shallow reduced tillage presented higher *S*. However, these effects were also site or year dependent within a given country. Climatic conditions had the most important impact on the decomposition rate *k* and the stabilization factor *S,* but also pH, treatment, crop types, SOC, C:N ratio and clay content were good predictors of the TBI parameters. Generally, the correlations with raw climatic variables such as precipitation and temperature were quite poor. Better relationships were found when nonlinearities due to interactions between climatic and edaphic conditions were accounted for. Our results highlights how a wide range of soil management practices affect TBI parameters jointly to soil and climatic conditions.

Data availability

Data can be provided by the authors upon request.

Author contribution

MRG, MB and TS wrote and prepared the manuscript draft; MB, TK and TS supervised and led the research; MB, OA, HS, JKF, AS, AS and TS developed the methodology; MRG and LM analyzed the data. All authors reviewed and edited the manuscript.

Competing interests

The contact author has declared that none of the authors has any competing interests.

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Tables

Table 1. Experimental sites in Austria. Site acronym, location name, year of TBI measurements, category and type of LTE including its duration, main crop cultivated during the TBI measurements and management treatments

Site	Location	Year	Category[*]	Experiment[†]	Age	Crop	Treatments[§]
AT1	MUBIL	2014	CB	OA	11	Luzerne	GM, FW, FYM, BS
AT2	MUBIL	2015	CB	OA	12	Wheat	GM, FW, FYM, BS
AT3	Ritzlhof	2015	CB	IF-N & OA+N	24	Maize	0, 40, 90, 120 N CFW, CGM, CS, CSS+80N
AT4	Rottenhaus	2016	CB	CR with NPK	31	Maize	CRR, CRI
AT5	Rutzendorf	2015	CB	CR with NPK	34	Wheat	CRR, CRI
AT6	Rutzendorf	2016	CB	CR with NPK	35	Maize	CRR, CRI
AT7	Breitstetten	2015	SF	IF-N with PK	40	Wheat	60, 90, 120, 145 N
AT8	Breitstetten	2016	SF	IF-N with PK	41	Wheat	60, 90, 120, 145 N
AT9	Haringsee	2015	SF	IF-N with PK	40	Wheat	60, 90, 120, 145 N
AT10	Fuchsenbigl	2016	SF	IF-K with NP	61	Maize	0, 150, 300 K
AT11	Rottenhaus	2016	SF	IF-K with NP	63	Maize	0, 150, 300 K
AT12	Haringsee	2016	SF	IF-N without PK	41	Maize	0, 60, 120, 180 N
AT13	Zinsenhof	2016	SF	IF-N without PK	41	Maize	0, 60, 120, 180 N
AT14	Zissersdorf	2016	SF	IF-N without PK	41	Maize	0, 60, 120, 180 N
AT15	Fuchsenbigl	2015	TS	TS	27	Maize	CT, SRT, DRT
AT16	Fuchsenbigl	2016	TS	TS	28	Wheat	CT, SRT, DRT

[*] CB: carbon balance practices; SF: soil fertility; TS: tillage systems.

[†] OA: organic amendments; IF-N & OA+N: N inorganic fertilization and organic amendments plus mineral N; CR & IF-P with NK: crop residues and P inorganic fertilization with 90 and 40 kg ha^{-1} of N and K, respectively; IF-N with PK: N inorganic fertilization with 55 and 180 kg ha^{-1} of P and K, respectively; IF-N without PK: N inorganic fertilization; IF-K with NP: K inorganic fertilization with 40 and 120 kg ha^{-1} of P and K; TS: tillage system.

[§] GM: green manure; FW: municipal compost and green manure; FYM: farmyard manure; BS: biogas slurry; CFW: compost food waste with 175 kg N ha^{-1}; CGM: compost green manure with 175 kg N ha^{-1}; CS: compost slurry with 175 kg N ha^{-1}; CSS: compost sewage sludge with 175 kg N ha^{-1}; CRR: crop residues removed; CRI: crop residues incorporated; CT: conventional tillage; SRT: shallow reduced tillage; DRT: deep reduced tillage.

Table 2. Experimental sites in Sweden. Site acronym, location name, year of TBI measurements conducted in 2016, type of LTE including its duration, main crop cultivated during the TBI measurements and management treatments

Site	Name	Experiment[*]	Age	Crop	Treatments[†]
SE1	Börgeby	CMP	59	Sugar beet	FYM/NPK
				Sugar beet	0 FYM/0 NPK
SE2	Ekebo	CMP	59	Sugar beet	FYM/NPK
				Sugar beet	0 FYM/0 NPK
SE3	Högåsa	CMP	50	Ley production year	FYM/NPK
				Ley production year	FYM/0 NPK
				Spring oilseed	0 FYM/NPK
				Spring oilseed	0 FYM/0 NPK
SE4	Kungsängen	CMP	53	Oat	FYM/NPK
				Oat	0 FYM/0 NPK
SE5	Röbacksdalen	CMP	47	Barley	FYM/NPK
				Barley	FYM/0 NPK
SE6	Vreta Kloster	CMP	50	Ley production year	FYM/NPK
				Ley production year	FYM/0 NPK
				Spring oilseed	0 FYM/NPK
				Spring oilseed	0 FYM/0 NPK
SE7	Lanna	ROT	35	Oat	SC
				Ley production year	L
SE8	Lönnstorp	ROT	36	Barley	SC
				Ley establish. year	L
SE9	Röbacksdalen	ROT	36	Barley	SC
				Ley establish. year	L
SE10	Säby	ROT	46	Wheat	SC
				Ley establish. year	L
SE11	Lanna	TS	34	Winter wheat	CT, DS
SE12	Säby	TS	11	Barley	CT, SRT, DRT, DS
SE13	Ultuna	TS	19	Barley	CT, DRT

[*] CMP: combined management practices; ROT: rotation systems; TS: tillage systems.

[†] FYM/NPK: maximum amount of farmyard manure and maximum doses of NPK; 0 FYM/0 NPK: no manure and no NPK application; FYM/0 NPK: maximum amount of farmyard manure and no NPK application; 0 FYM/NPK: no manure and maximum doses of NPK; SC: spring cereal; L: ley; CT: conventional tillage; DS: direct seeding; SRT: shallow reduced tillage; DRT: deep reduced tillage.

Table 3. Annual climatic characteristics for the Austrian and Swedish sites during the entire year of measurements and only during the TBI period (days) corresponding to the period between the date of placement and the last retrieval date of the tea bags.

Site	TBI period	TAP	TP_{TBI}	MAT	MAT_{TBI}	PET	PET_{TBI}	AI	AI_{TBI}
	days	mm		ºC		mm			
Austria									
AT1	99	756	294	12.5	20.3	811.4	414.0	1.07	1.4
AT2	61	516	65	12.6	18.8	605.1	141.3	1.17	2.2
AT3	80	622	161	12.2	20.0	682.9	295.1	1.09	1.8
AT4 & 11	84	939	384	11.2	20.5	763.4	306.1	0.81	0.8
AT5	63	516	82	12.6	19.4	605.1	148.6	1.17	1.8
AT6	96	735	226	12.0	21.6	854.2	394.0	1.16	1.7
AT7	59	516	81	12.6	19.3	605.1	137.4	1.17	1.7
AT8	84	735	240	12.0	17.1	854.2	334.2	1.16	1.4
AT9	60	516	81	12.6	19.2	605.1	139.6	1.17	1.7
AT10 & 16	92	735	205	12.0	21.4	851.2	378.9	1.15	1.8
AT12	87	735	203	12.0	21.8	854.2	367.4	1.16	1.8
AT13	84	939	327	11.2	20.5	763.4	304.3	0.81	0.9
AT14	82	735	154	12.0	21.3	854.2	319.6	1.16	2.1
AT15	81	516	103	12.6	22.5	605.1	212.9	1.17	2.1
Sweden									
SE1	92	436	141	9.2	16.7	444.6	235.1	1.02	1.7
SE2	91	681	270	9.0	16.5	578.5	297.8	0.85	1.1
SE3	91	458	142	7.9	13.3	618.0	306.4	1.35	2.1
SE4	93	429	126	7.1	15.3	640.0	367.0	1.48	2.9
SE5 & 9	92	526	164	4.1	14.3	396.2	218.2	0.75	1.3
SE6	91	433	158	7.9	13.3	617.5	301.8	1.16	1.9
SE7 & 11	91	412	115	7.4	12.7	514.2	276.4	1.3	2.4
SE8	90	610	224	9.4	14.4	500.2	265.7	0.82	1.2
SE10 & 12	91	429	126	7.1	15.3	358.5	155.7	0.84	1.2
SE13	92	429	126	7.1	15.3	639.1	377.8	1.5	3.0

TAP: total annual precipitation; TP_{TBI}: total precipitation during TBI period; MAT: mean annual temperature; MAT_{TBI}: mean temperature during TBI period; PET: total annual potential evapotranspiration; PET_{TBI}: potential evapotranspiration during TBI period; AI: annual aridity index (PET divided by TAP); AI_{TBI}: aridity index during TBI period.

Table 4 – Mean values of decomposition rate (*k*) and stabilization factor (*S*) for the TBI approach after the incubation period.

	Mean TBI parameters	
	k	*S*
Sweden	0.0160 ± 0.01	0.247 ± 0.14
Austria	0.0115 ± 0.004	0.228 ± 0.11

Figures

FIGURE 1

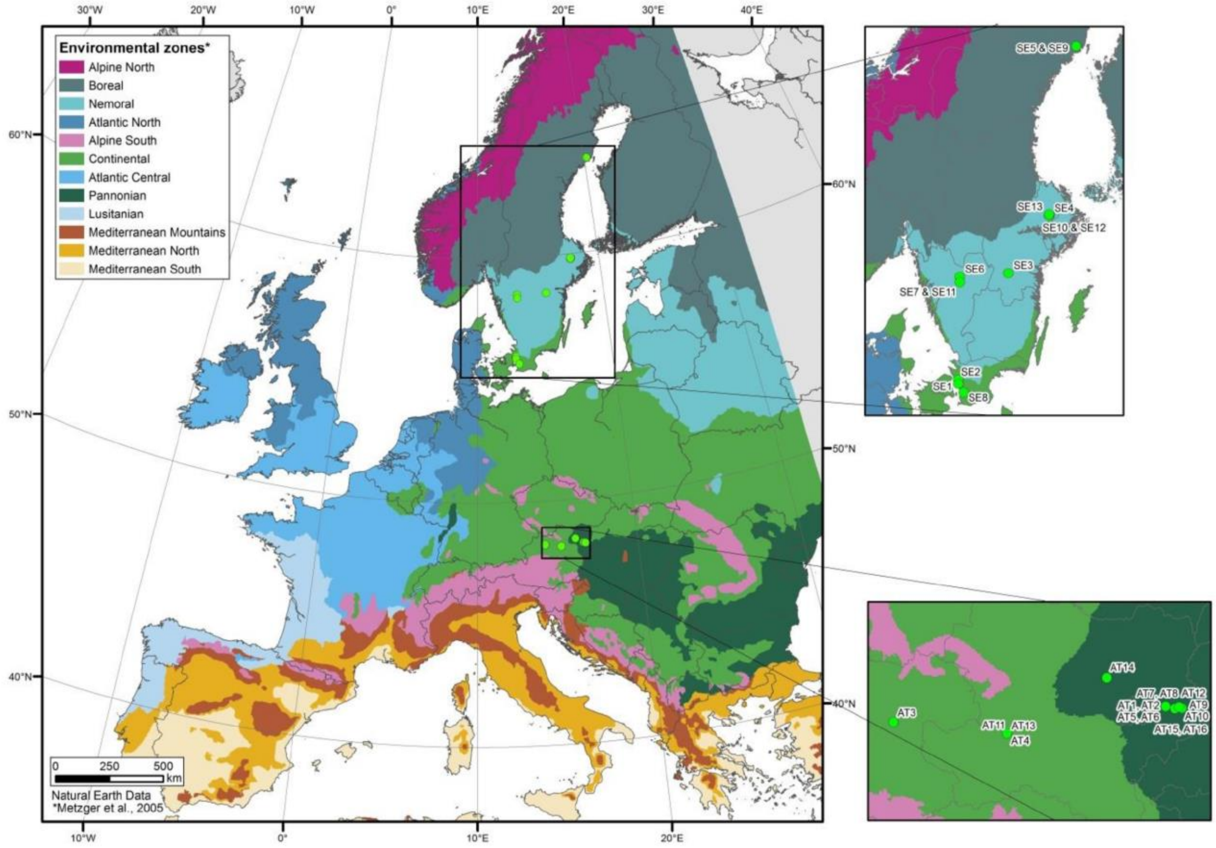
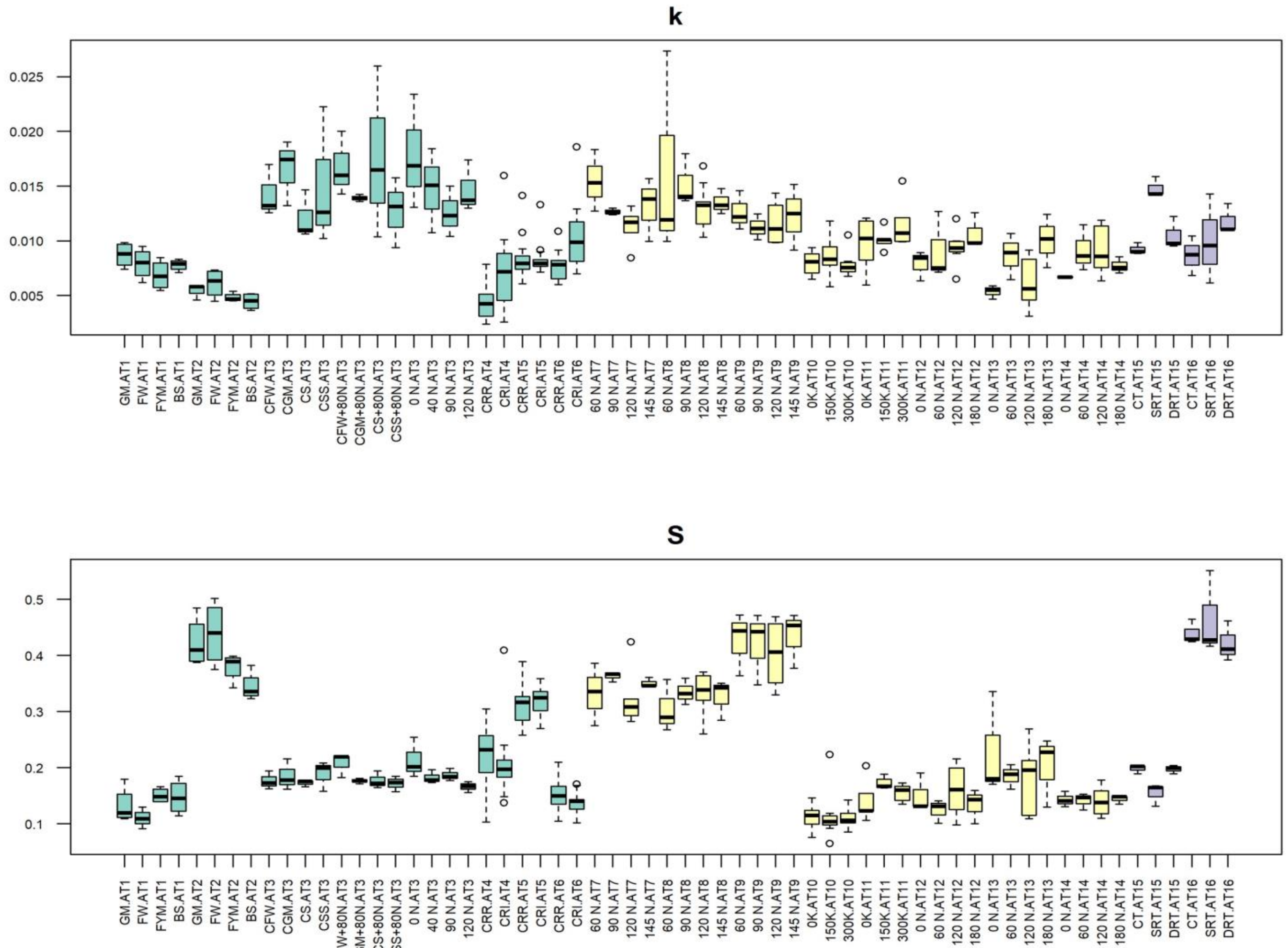


FIGURE 2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BS.AT2
CFW.AT3
CGM.AT3
CS.AT3
CSS.AT3
CFW+80N.AT3
CGM+80N.AT3
CS+80N.AT3
CSS+80N.AT3
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CRR.AT4
CRI.AT4
CRR.AT5
CRI.AT5
CRR.AT6
CRI.AT6
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120 N.AT13
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DRT.AT15
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DRT.AT16
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145 N.AT9
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150K.AT10
300K.AT10
0K.AT11
150K.AT11
300K.AT11
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60 N.AT12
120 N.AT12
180 N.AT12
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180 N.AT13
0 N.AT14
60 N.AT14
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CT.AT16
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DRT.AT16

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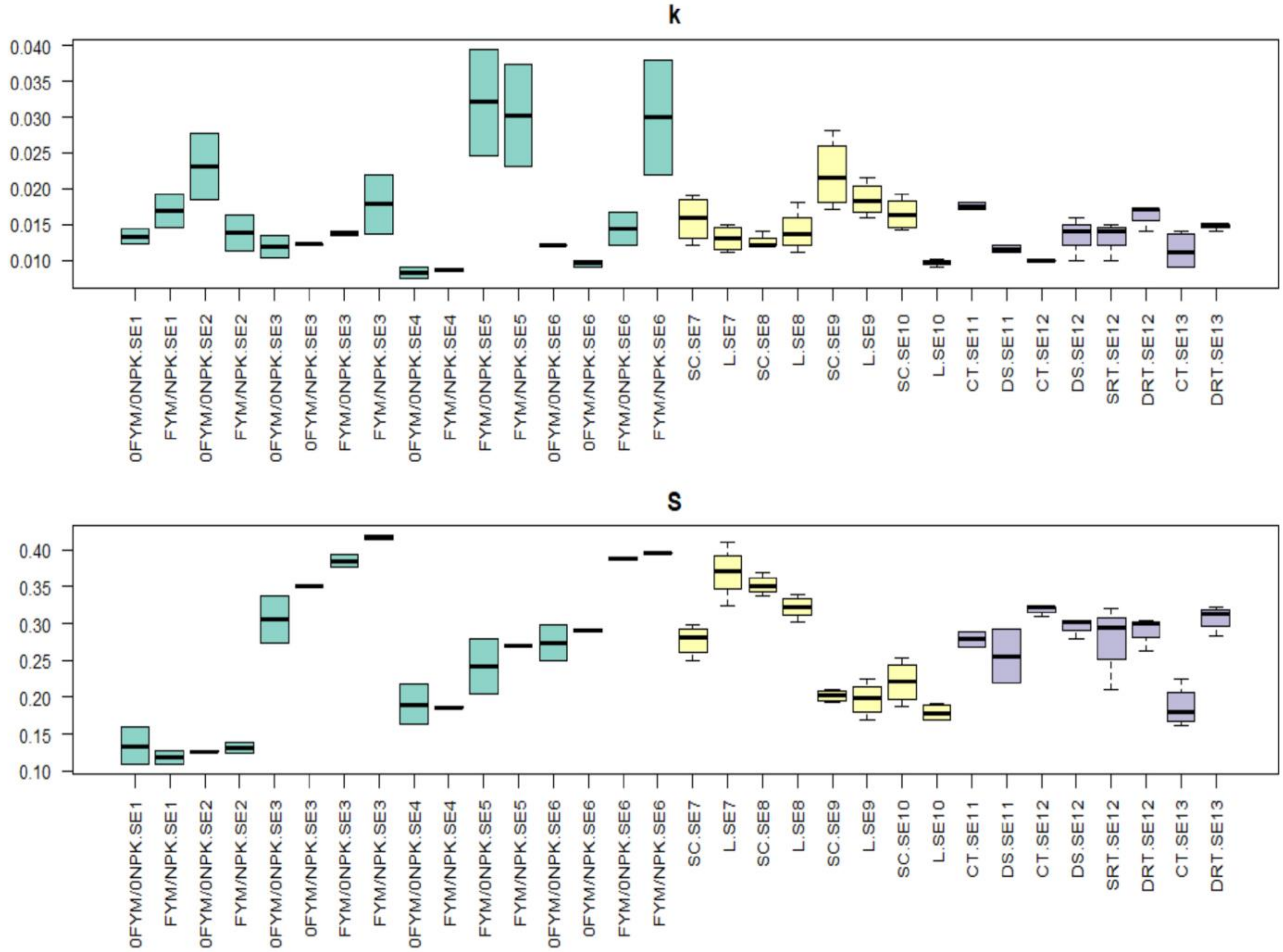
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SC.SE10
L.SE10
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CT.SE12
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CT.SE13
DRT.SE13
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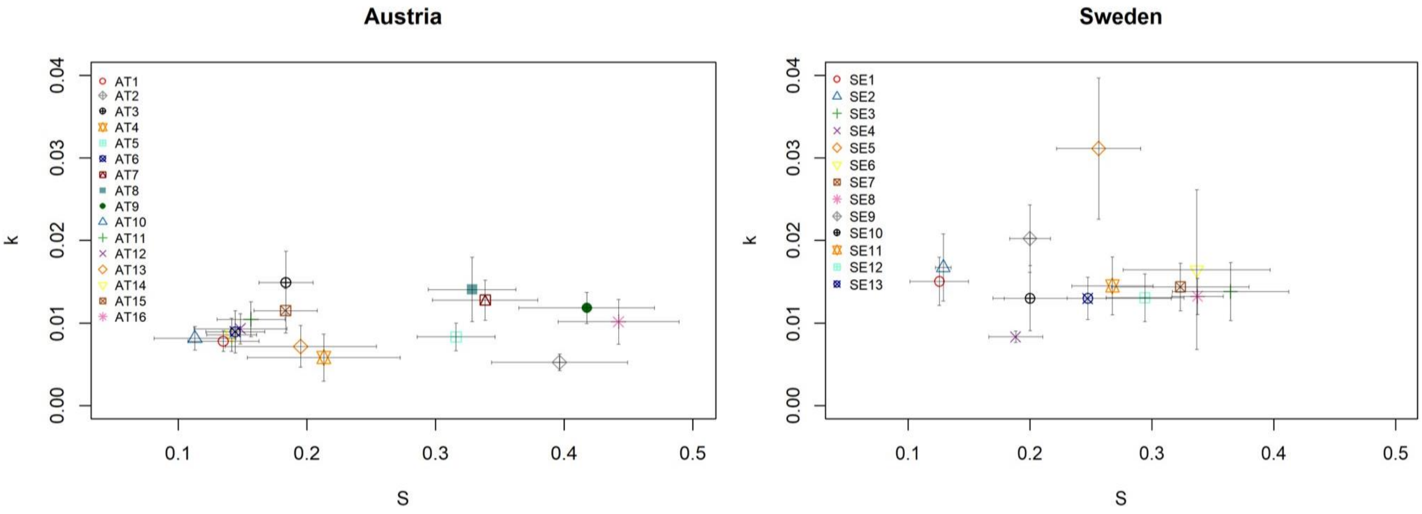
FIGURE 4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AT1
AT2
AT3
AT4
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AT6
AT7
AT8
AT9
AT10
AT11
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AT14
AT15
AT16
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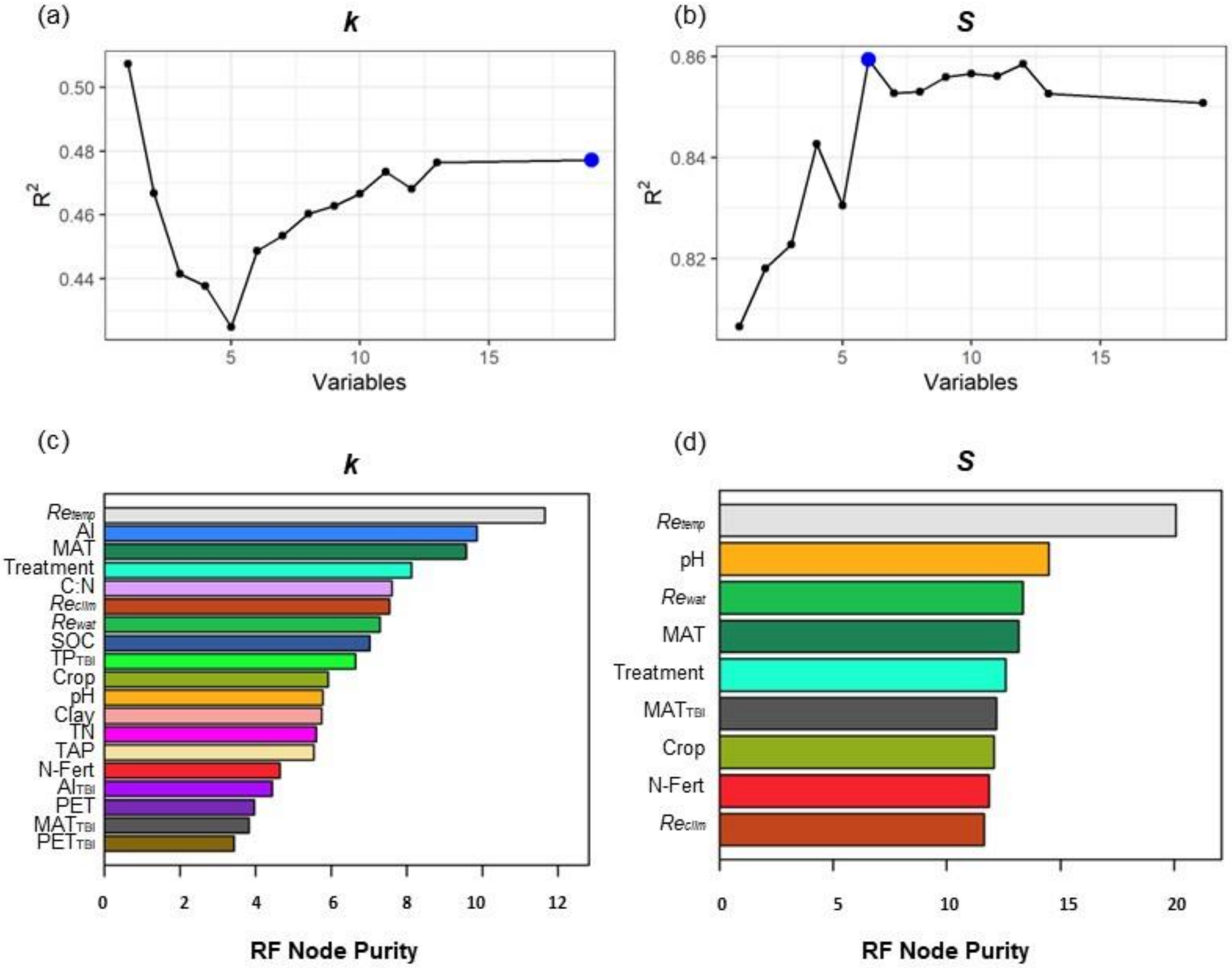
SE13

FIGURE 5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(b)
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Variables
(c)
k
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AI
MAT
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C:N
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12
RF Node Purity
(d)
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FIGURE 6

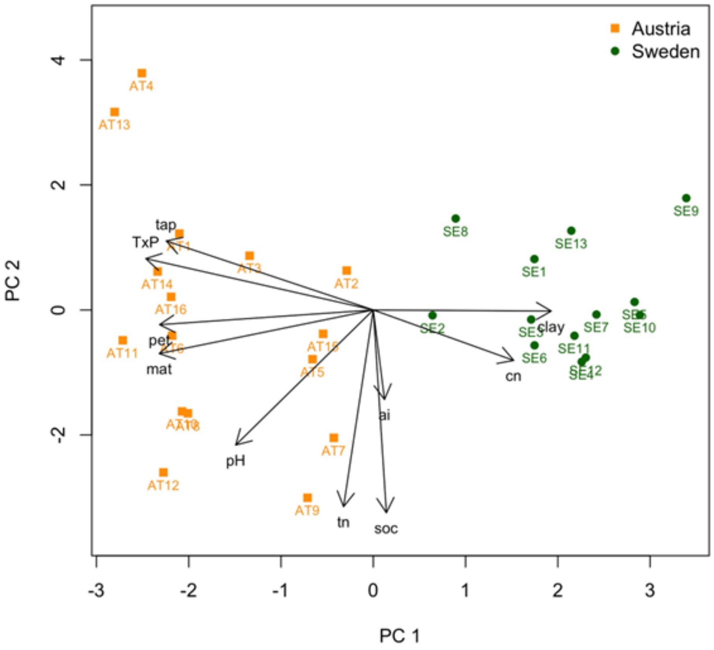

Figure caption

Figure 1 - Location and environmental zone of the Austrian and Swedish sites.

Figure 2 - Average decomposition rate (*k*) and stabilization factor (*S*) for each treatment and site in Austria. The extents of the box indicate 25th and 75th percentiles, and the lines represent the 50th percentile. Whiskers represent the 10th and 90th percentiles and outliers are given as open symbols. Green boxes: Carbon balance (CB) experiment; yellow boxes: Soil fertility (SF) experiment; purple boxes: tillage systems (TS) experiment. Site AT1 shows results from 2014. Sites AT2, AT3, AT5, AT7, AT9, and AT15 show results from 2015. And sites AT4, AT6, AT8, AT10, AT11, AT12, AT13, AT14, and AT16 show results from 2016.

Figure 3 - Average decomposition rate (*k*) and stabilization factor (*S*) for each site and treatment in Sweden. The extents of the box indicate 25th and 75th percentiles, and the lines represent the 50th percentile. Whiskers represent the 10th and 90th percentiles. Green boxes: combined management practices (CMP) experiment; yellow boxes: rotation (ROT) experiment; purple boxes: tillage systems (TS) experiment.

Figure 4 - Distribution of the mean decomposition rate constant (*k*) and the stabilization factor (*S*) for each site in Austria and Sweden. Errors bars represent standard deviation.

Figure 5 - a and b) Variables selection procedure to identify the optimal number of variables to explain the variance of *k* and *S* with a Random Forest model. The blue point represents the optimal model. c and d) Relative importance of the variables used by each optimized Random Forest model to predict the variance in the *k* and *S* parameters in Austria and Sweden jointly. The higher the Node purity, the higher the importance of such variable.

Figure 6 - Principal component analysis showing how the sites in Austria and Sweden differ based on the variables. PC1 and PC2 are the first two components, explaining most variance. The loadings (black arrows) are the weight of each variable in defining each principal component. The size of the arrows can tell how much they contribute defining this space, while the direction is their contribution on each axis. Tap: total annual precipitation; TxP: temperature x precipitation factor; pet: potential evapotranspiration; mat: mean annual temperature; tn: total soil nitrogen; soc: total soil organic carbon; cn: soil C:N ratio; ai: aridity index.