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- 1 Evaluating the Tea Bag Index approach for different management practices in
- 2 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 ~60 and 90 days in 16 Austrian and 13 Swedish sites. The treatments at Austrian LTEs focused on mineral and organic fertilization, tillage systems and crop residues management, whereas the Swedish LTEs addressed cropping systems, mineral fertilization and tillage systems. The results showed that in Austria, decomposition differed more between sites than between treatments for the same experiment category. Incorporation of crop residues and high N fertilization increased k. Minimum tillage had significantly higher k compared to reduced and conventional tillage. In Sweden, litter decomposition differed more between treatments than between sites. Fertilized plots showed higher S than non-fertilized and high N fertilization had the highest k. Growing spring cereal lead to higher k than forage. Random Forest regressions 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



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dynamics. Thus, the TBI approach may be suitable to apply in a more large-scale network on LTEs

48 for evaluating decomposition dynamics 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 fertilization, 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 newly added litter resulting from microbial activity (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;





70 Gholz et al., 2000). Decomposition is an extended process, therefore long-term field experiments 71 (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., 72 73 2012; Bergkvist and Öborn, 2011). Experiments determining litter mass loss over time in situ are 74 also important for understanding SOC dynamics, nutrient cycling and colonization by soil biota 75 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, 76 77 and retrieving them successively at different intervals (Kampichler and Bruckner, 2009; Burgess 78 et al., 2002; Bocock and Gilbert, 1957). These studies are not always comparable because they are 79 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, 80 81 2009). Keuskamp et al. (2013) developed therefore a low-cost and time-efficient methodology called Tea 82 83 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 84 85 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 86 incubation period of ca 90-days in the soil. The TBI approach is particularly useful for assessing 87 88 geographical differences in decomposition dynamics because results are directly comparable 89 across sites, varying only with local edaphic and seasonal environmental conditions (Keuskamp et al., 2013). 90 91 In several studies, the TBI has been used as an indicator for biological (Sandén et al., 2021; 92 Costantini et al., 2018) and microbial activity (Daebeler et al., 2022; Treharne et al., 2019; Tóth et https://doi.org/10.5194/egusphere-2023-1229 Preprint. Discussion started: 26 June 2023 © Author(s) 2023. CC BY 4.0 License.



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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. Most studies have been using the TBI approach for different forest and grassland ecosystems (Djukic et al., 2018) or urban soils (Pino et al., 2021). Only a few 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) 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. 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. The treatments covered management practices such as organic amendments, crop rotations, aboveground crop residue handling, mineral fertilization, and tillage. Our objectives were to evaluate: (i) if the TBI k and S parameters are sensible enough to detect between different management practices for agroecosystems; (ii) to quantify the effect of management practices on k and S; (iii) and to identify the most important local climate and/or soil properties affecting litter decomposition in Austria and Sweden.

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Materials and Methods

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Study sites

115 Austria





116 We used sixteen Austrian (AT) sites, by selecting contrasting treatments from three different 117 categories of LTEs where the management practices had been in place for 11 to 63 years (Table 1). TBI measurements were made in 2014, 2015 and 2016. Measurements sometimes took place 118 in more than one year at the same LTE (e.g., MUBIL), and the sites were abbreviated as AT1 to 119 AT16. Six experiment categories involved C balance practices (CB; AT1 to AT6), eight sites were 120 121 studying soil fertility (SF; AT7 to AT14) and two sites examined tillage systems (TS; AT15 to AT16). The sites are located in several agricultural areas across the country (Fig. 1), with diverse 122 soil textures and variable crop types (Table 1) and climatic characteristics (Table 2) during the 123 years of TBI measurements. More details for some of the sites are available in specific 124 publications: AT3 (Spiegel et al., 2018; Lehtinen et al., 2017; Tatzber et al., 2015; Aichberger and 125 Söllinger, 2009), AT4 to 6 (Spiegel et al., 2018; Lehtinen et al., 2014), AT15 and AT16 (Tatzber 126 127 et al., 2015; Spiegel et al., 2007). In addition, Sandén et al. (2018) puts the Austrian LTEs in the context of other European LTEs. 128 129 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), 130 131 ii) to investigate the effects of compost amendments on soil and crops (AT3), and iii) to compare 132 crop residue incorporation and removal (AT4-AT6). Compost amendment LTE and crop residue incorporation LTEs included also mineral fertilization, whereas AT1 and AT2 only focused on 133 134 different organic fertilization treatments. The eight soil fertility LTEs (AT7-AT14) were all focusing on the effect of mineral fertilization 135 136 on soil and crop properties. In most cases, treatments studied different amounts of mineral nitrogen 137 fertilization, whereas AT9 and AT12 also investigated the effect of different amounts of K





138 fertilization. Nitrogen fertilization was applied in four stages and potassium in three stages, 139 according to Austrian guidelines for fertilization (BMLFUW, 2017). For the tillage system experiments, conventional tillage (CT) was compared to reduced tillage (RT) 140 and minimum tillage (MT). Regular mouldboard ploughing to 25-30 cm soil depth was applied in 141 142 CT treatment, whereas cultivator in autumn to a depth of 15-20 cm was used in RT treatment and 143 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. 144 Fertilization was crop specific according to the Austrian guidelines for fertilization (BMLFUW, 145 146 2017). 147 Sweden 148 149 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 (Table 150 151 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, SE1 to SE6), four 152 153 studying the effect of rotations (ROT, SE7 to SE10) and three sites with tillage systems (TS, SE11 154 to SE13). 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 155 156 year of TBI measurements. Bergkvist and Öborn (2011) give a general description of all these LTEs, more details on the sites with combined management practices is given by Carlgren and 157 Mattson (2001), and for tillage systems by Arvidsson et al. (2014), while Poeplau et al. (2015) 158 159 provide some more insight on the rotation experiments.

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The initial purpose of the LTEs with combined management practices 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 perennial grass-clover leys and receives 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 (NPK). PK applications in all the treatments we selected were aimed 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 in these CMP, 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. We were comparing extreme treatments representing two rotations from three LTEs 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 tillage experiments, the conventional tillage (CT) and direct seeding (DS) treatments were the same for all sites, consisting of inversion ploughing 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 sinensin*; EAN: 8722700055525) has high cellulose content, higher soluble fraction, and lower C:N ratio; while rooibos tea (*Aspalanthus 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° 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



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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, only one site used successive retrieval dates (AT16), in which four bags of each tea were used and placed side-by-side at a distance of 2 to 3 cm, in order to keep as similar soil characteristics as possible. In this case, the tea bags collecting occurred after 16, 26, 62 and 91 days. For the other Austrian sites, there was only one collecting, and the TBI incubation period from placement to last retrieval averaged 80±13 days (Table 3). After collecting, the tea bags were cleaned of soil and roots and oven-dried at 70°C for 48 hours. 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 but all the sites used successive retrieval dates. As in Austria, four bags of each tea were used per experimental unit for each retrieval date, 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 tea bags were collected after four different time periods of ~15, 30, 60 and 90 days. The mean TBI incubation period from placement to last retrieval date averaged 91±1 day (Table 3). To quantify soil contamination, the ash content was determined for each of the four retrieval dates (i.e., both for green and rooibos tea on mixed samples of the four replicates) in a muffle oven at 550°C for 16 hours. After measuring the remaining dry matter, the decomposition rate (k) and stabilization factor (S) were calculated according to Keuskamp et al. (2013). 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 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).



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soil organic C (TOC) concentrations were analyzed by dry combustion in a LECO RC-612 TruMac 229 CN (LECO Corp., St. Joseph, MI, United States) at 650°C (ÖNORM L1080). Total N (Ntot) was 230 231 determined according to ÖNORM L1095 with elemental analysis using a CNS (carbon, nitrogen, 232 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. Clay 233 234 content, C content, C:N ratio, and pH from each site in both countries are shown in Table S1 235 (Supplementary material). 236 237 Data analysis 238 Analysis of variance was performed to analyze treatment and site effects on k and S, followed by the Tukey's test (p < 0.05) using R software version 4.2.2. Interactions between site and treatment 239 240 were considered. We used a climate-dependent soil biological activity scaling function (Re_{clim}), which is included in 241 the ICBM SOC model (Andrén and Kätterer, 1997) for adjusting the decomposition rates of SOC 242 243 pools (Andrén et al. 2004; 2007). This function is integrating the effect of climate, soil and crop properties by calculating the product of soil temperature (Re_{temp}) and relative water content (Re_{wat}) 244 245 in the arable layer. These two variables are derived from soil temperature and moisture response

functions expressing the activity of decomposers and their relative effect on decomposition

kinetics. The Retemp 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 potential evapotranspiration (PET). In addition

to air temperature and leaf area index, calculations of Re_{wat} also involve the use of daily climatic

For Austrian soils, pH was measured electrochemically (pH/mV Pocket Meter pH 340i, WTW,

Weilheim, Germany) in 0.01 M CaCl₂ at a soil-to-solution ratio of 1:5 (ÖNORM L1083). Total



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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). We calculated simple correlation between variables 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 utilizing treatment and crop variables (including N fertilization) 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 kinetic parameters k and S (considering data from measurements only at 60 days and only at 90 days, or all measurements from 60 and 90 days combined):



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 $k_n f(TN, N, SOC, PET_{TBI}, PET, cr, TP_{TBI}, TAP, tr, cl, CN, pH, MAT, MAT_{TBI}, AI, AI_{TBI}, Re_{clim}, Re_{wat}, Re_{temp})$ 275 $S_n f(TN, N, SOC, PET_{TBI}, PET, cr, TP_{TBI}, TAP, tr, cl, CN, pH, MAT, MAT_{TBI}, AI, AI_{TBI}, Re_{clim}, Re_{wat}, Re_{temp})$ 276 277 where the subscript n denotes the grouping based on how long period the tea bags were in the soil 278 (i.e., for 60 or 90 days, or both periods combined). Soil variables (continuous) were TN (total N g 279 kg⁻¹), SOC (g kg⁻¹), CN (C:N ratio), cl (clay content g 100 g⁻¹) and pH. Categorical variables were 280 281 N (N fertilization factor with 4 levels), cr (a crop factor, e.g., barley, ley (establishment), ley (production), oat, spring oilseeds, sugar beet and winter wheat) and tr (a treatment factor with 30 282 levels). The climatic variables (PET_{TBI}, PET, TP_{TBI}, TAP, MAT_{TBI}, MAT, AI_{TBI} and AI) are as 283 defined in Table 3. The climate response variables Re_{clim} , Re_{wat} , Re_{temp} are as described above. 284 Since many of variables in our model are likely to be correlated and carry similar information, we 285 applied the recursive feature elimination algorithm implemented in the caret R package by Kuhn 286 et al. (2016), which assess in subsequent iterations the optimal set of predicting variables (features) 287 to be utilized by the RF model. The procedure starts by fitting a RF model with all variables, 288 289 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 R²), 290 selecting the set with the best model fitness. The selected models were used to compute the 291 292 variables' relative importance.





294	Results

295 Effect of management practices

296 Austria

Both the TBI parameters k and S varied between treatments and sites in Austria, and even between 297 years at the same site within the C balance category (Fig. 2 and Table S3). In general, all treatments 298 299 in AT1 with a lucerne crop under wetter conditions (2014) presented higher k and lower S than AT2 with a wheat crop under dryer conditions (2015), and the FW treatment had the highest S in 300 2015. The AT3 site did not present significant differences between the treatments. Treatment CRI 301 302 had higher k than the CRR treatments at the AT4 and AT6 sites, and AT6 presented a higher k than 303 at AT4. Comparing years for the same experiment type, AT5 (2015) had higher S than AT6 (2016). For the soil fertility experiment category (Table 1), AT12 had the highest k and AT13 had the 304 highest S. Sites receiving NPK fertilization (AT7, AT8 and AT9) had higher k and S even at 305 different N doses than at the AT12, AT13 and AT14 sites receiving only N (i.e., without P and K). 306 307 Stabilization was significantly higher in AT9 than in AT7. For K trials, AT11 presented significantly higher k and S than AT10. Regarding sites receiving N addition only (AT12, AT13, 308 309 and AT14), maximum doses (180 kg N) presented the highest k (0.0095), and no N addition had 310 the lowest k (0.0066) (Fig. 2 and Table S2). Regarding the tillage system experiment category at the Fuchsenbigl LTE (Table 1), k was 311 312 significantly higher in SRT and S was higher in DRT in 2015 (AT15), but no significant differences between treatments were found in 2016 (AT16). Site AT16 had significantly higher S than AT15 313 314 (Fig. 2. and Table S2).

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Sweden





317 At the Swedish sites (Table 2), the 90 days TBI measurements for the combined management 318 practices experiment category showed that both k and S were significantly higher for the FYM/NPK treatments (Fig. 3, Table S3) compared with the control treatments (0 FYM/0 NPK). 319 Comparing sites, SE5 and SE6 had highest k and SE4 had the lowest k, while SE3 presented the 320 321 highest S followed by SE6, SE5 and SE4, S was lowest for SE1 and SE2 (Table S3). 322 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 323 324 SE7, SE8 and SE10, whereas SE7 and SE8 had the highest *S*. 325 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 326 followed by SE11 and SE13. Sites did not show significant differences for k. 327 328 Comparing tillage system experiments in Austria and Sweden (2016) for sites (AT16, SE11, SE12 and SE13) and treatments (CT, SRT and DRT), DRT had higher k and SRT had higher S. The 329 330 Austrian site presented the lowest k and the highest S compared to the Swedish sites, which did 331 not present significant differences among them (Table S3). 332 Mean k by site in Austria varied between 0.0053 and 0.0149, and mean S varied between 0.113 and 0.442 (Table S3). Mean k by site in Sweden was between 0.0084 and 0.0311, and mean S was 333 between 0.125 and 0.365 (Table S3). All values for k and S were within the range of the previous 334 335 global TBI investigation (0.005-0.04 for k; and 0.05-0.55 for S) by Sandén et al. (2020). The mean values of the TBI decomposition rate and the stabilization factor were both higher at 60 336 days than at 90 days, in Austria as well as in Sweden (Table 4). After 90 days of incubation, mean 337 338 k was higher in Sweden and mean S was higher in Austria. Applying a decomposition model to the series of data from successive retrieval dates (i.e., all the Swedish sites and the AT16 site in 339



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Austria) on the remaining dry matter over time showed faster decomposition of Green compared to Rooibos tea, which is in agreement with the TBI concept (Fig. S1). Whereas the decomposition curve for Rooibos kept decreasing after 90 days, that of green tea did not decrease any further after about 60 days. The variability between sites in dry matter loss over time was higher for Green than for Rooibos tea. The field application of the TBI found a clear discrimination of both k and S between agroecosystems in Austria and Sweden after the incubation period (Fig. 4). Influence of climate and soil properties Using the combined dataset for the 90 days TBI period 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, Reclim and Retemp. After the 60 days TBI period, Austria and Sweden combined presented significant negative correlation between k and MAT, PET, AI, T x P factor, pH and clay content, and a positive correlation with TAP and C:N ratio. Stabilization correlated negatively with the C:N ratio. The variable selection procedure with the random forest models (Fig. 5) identified fewer variables explaining k and S values for the combined dataset. When considering only the 90 days subset, the variables explaining k increased, but the overall predicting power of the model decreased substantially. A similar pattern, but less strong, was noticed for the 60 days subset. More than 70% of the variance of k for the combined dataset (i.e., 60 and 90 days TBI period) was accounted for by climatic variables only (Fig. 6), with Rewat and Retemp ranking the highest followed by Reclim, AI and MAT, according to the optimized random forest model. On the contrary, S was influenced by much more factors, again with climate-related variables leading the ranking but https://doi.org/10.5194/egusphere-2023-1229 Preprint. Discussion started: 26 June 2023 © Author(s) 2023. CC BY 4.0 License.





including also many edaphic characteristics, such as pH, SOC, clay and nitrogen content and the C:N ratio, as well as agronomic variables such as treatment, crop and N fertilization. The rankings when using the two subsets of data separately (i.e., 60, and 90 days TBI periods) were less relevant since the predictive power of the model decreased compared to when using the combined dataset. This was particularly true for k, where the overall cumulated node purity also decreased substantially compared with the combined dataset.



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Discussion

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 6). This is in contrary 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 on early-stage litter decomposition in a temperate biome. In the C balance trials in Austria, soils receiving green manure + municipal compost (FW) had higher S than soil receiving biogas slurry (BS) 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 CRI 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 PET resulting in lower AI at the AT4 site (Table 3 and Table S1). 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



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lower clay content, lower precipitation resulting in higher PET 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. In the tillage system experiment at Fuchsenbigl in Austria in 2015 (AT15) and 2016 (AT16), the shallow tillage (SRT) showed significantly higher k than DRT and CT, but only in 2015, indicating that shallow soil tillage stimulated decomposition that particular year. 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 lower AI_{TBI} during the TBI period. In the Swedish combined management practices trials, soil treatments receiving organic and mineral fertilization 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 PET and AI, resulting in more moisture; SE6 had also high S, due to high clay content and low PET. Site SE3 had high S, which could be related to





415 a higher C:N ratio, as suggested by Althuizen et al. (2018) that C:N ratio is positively correlated to S. SE1 and SE2 had lower S than SE3 despite similar climatic conditions, which probably was 416 related to the crops growing in these treatments (i.e., sugar beet in the SE1 and SE2 and 417 grass/clover lev in SE3), which have different effects on soil temperature and moisture. 418 419 In the Swedish rotation system trials, spring cereal (SC) had higher k than ley (Table S3). Site SE9 420 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. The highest S were found in SE7 and SE8, in 421 which the former presented high clay and SOC content, and SE8 had high precipitation and low 422 PET. 423 For the tillage system treatments in Sweden, similar to the Austrian sites, the conventional tillage 424 presented the lowest k, and also lowest S. Even when comparing tillage systems in Sweden and 425 426 Austria jointly (Table S3) we could notice that conventional tillage also presented the lowest k, while DRT the highest. 427 428 The mean k was higher in Sweden, while the mean S was higher in Austria (Table 4, Fig. 4). In general, the variation in k values were lower in Austria, while the variation in S were lower in 429 430 Sweden. It is possible that the ash correction, that was made for the Swedish but not the Austrian sites, may partly explain this difference. Indeed, the average mass loss after 90 days at the Swedish 431 sites for Green and Rooibos tea was higher with about 60 and 30%, respectively, whereas it was 432 433 only about 45 and 15% for the Austrian sites (data not shown). When recovering litter dry matter from the soil, soil-contamination are often not negligible. In our study, the ash-content determined 434 435 on the Green and Rooibos tea bags for the Swedish site represented 15±6 and 10±4 %, respectively 436 (data not shown).



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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. Despite that many studies have showed a positive correlation between precipitation and decomposition rates (Pimentel et al., 2019; García Palacios et al., 2016), precipitation did not have a huge impact in our study according the random forest analysis. On the other hand, Re_{wat} showed great importance (Fig. 6). 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 (AI) was lower. AI was identified by the random forest regression model being an important variable affecting the rate of decomposition (Fig. 6). Soils from more arid and warmer sites are associated with lower SOC (Kerr and Ochsner, 2020; Ontl and Schulte, 2012). With increasing aridity, the biological processes that drive C and N inputs and fluxes in ecosystems may be impaired, which may result in decreasing soil C and N stocks (Jiao et al., 2016; Reynolds et al., 2007). The random forest models showed that the decomposition rate k was mostly affected by climate, in particular when considering the TBI periods combined (Fig. 6a). The lower predictive power of the models when considering the 60 and the 90 days TBI periods separately can explain the higher number of variables considered, due to less defined effects to be identified by the model. This is suggested also by the decrease in the overall node purity of the models using only 60, or 90 days





461 data to explain k. When using the combined dataset, the model was instead explaining a relatively large part of the variance ($R^2=0.735$) and with a much higher node purity, while employing very 462 few and only climatic-related parameters. 463 For practical reasons the teabags in our study were buried in the soil during the growing season, 464 465 corresponding to a period when the soil biological activity is highest (Bolinder et al., 2013). When 466 burying the teabags during the growing season, the difference in climate between sites are attenuated, in particular with respect to air temperature. For example, the MAT at the Swedish 467 most northerly (SE5 and SE9) and southerly (SE2) sites are 4.1 and 9.0 °C, respectively, whereas 468 corresponding mean air temperatures (MAT_{TBI}) during our study were 14.3 and 16.2 °C, 469 respectively. 470 The TBI S parameter was also dependent on climate. Indeed, the random forest model identified 471 472 climatic parameters as the main factors affecting S in both Austria and Sweden during all evaluated periods. In particular, Reclim and Retemp often showed significant negative correlations, which 473 474 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 475 476 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 477 478 is influenced by multiple factors that interact and change over time (Bradford et al., 2016), the 479 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 480 481 would not be detectable by linear approaches. 482 The stabilization factor S expresses the degree by which the labile fraction of the plant material is decomposed. Therefore, it is not surprising that more variables come into play to define it. In 483

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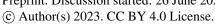




particular, we noticed the influence of edaphic factors, of which pH was the most important, but also SOC concentration, C:N ratio and clay were also identified as good predictors. In addition, agronomic factors were also influencing *S*, where soil management treatment and crop types were the most important. 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). Nevertheless, the impact of pH on SOC kinetics seems clear in our study with a maximum effect at around neutral pH (Liao et al., 2016).

Conclusion

Our results show that both 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 fertilization, 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 fertilization 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 SOC, C:N ratio and clay content were good predictors of the TBI





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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 bring knowledge and answers under how a wide range of soil management practices affect soil decomposition jointly to soil and climatic conditions. We recommend the TBI approach for further LTE studies evaluating soil decomposition dynamics. 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. Acknowledgements Financial support was provided by the Swedish Farmers' Foundation for Agricultural Research, grant number O-18-23-141. Part of this research has been done in the framework of the EJP SOIL that has received funding from the European Union's Horizon 2020 research and innovation programme: Grant agreement No 862695.





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792 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	СВ	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 120 kg N ha $^{-1}$; 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.



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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* Crop Treatments[†] Age Börgeby SE1 CMP Sugar beet FYM/NPK Sugar beet 0 FYM/0 NPK SE2 Ekebo CMP59 Sugar beet FYM/NPK Sugar beet 0 FYM/0 NPK SE3 Högåsa CMP 50 FYM/NPK Ley production year 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 47 CMP Barley FYM/NPK Barley FYM/0 NPK SE6 Vreta Kloster CMP50 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 SC Oat Ley production year L SE8 Lönnstorp ROT 36 Barley SC Ley establishment year L SE9 ROT Röbacksdalen 36 Barley SC Ley establishment year L SE10 Säby ROT 46 Wheat SCLey establishment year L SE11 Lanna TS 34 Winter wheat CT, DS SE12 Säby TS Barley CT, SRT, DRT, DS 11

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TS

Barley

CT, DRT

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SE13

Ultuna

^{*} 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	n	nm		°C	n	nm		
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 60 and 90 days of incubation period.

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Incubation	Mean TBI parameters				
incubation	k	S			
Sweden					
60 days	0.0160	0.296			
90 days	0.0152	0.267			
Austria					
60 days	0.0152	0.429			
90 days	0.0115	0.423			

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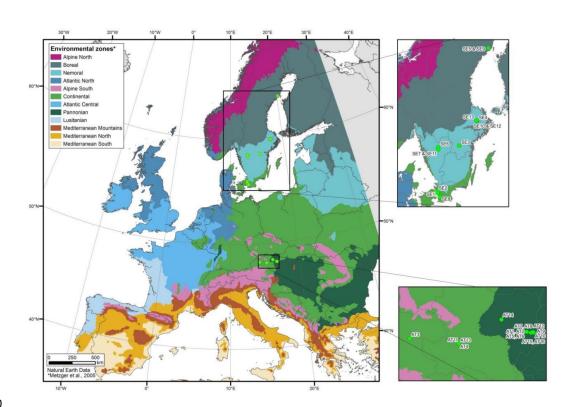




837 Figures

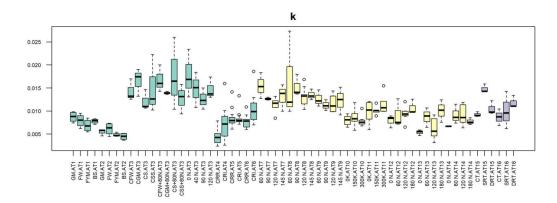
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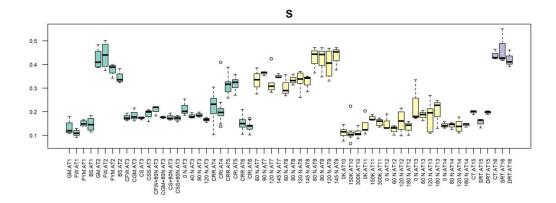
839 **FIGURE 1**







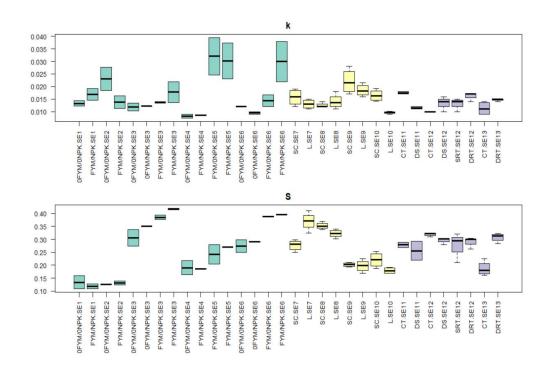








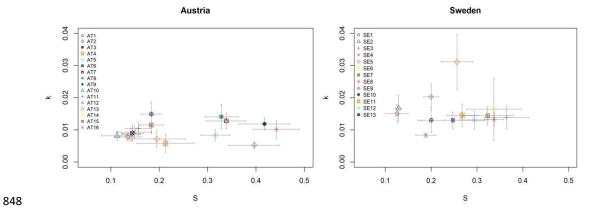
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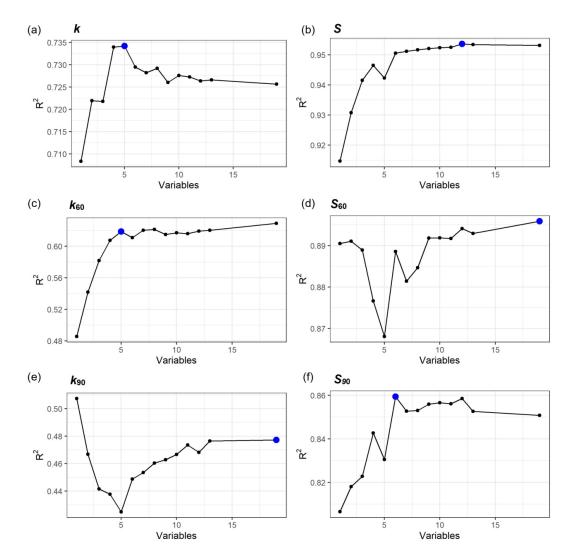
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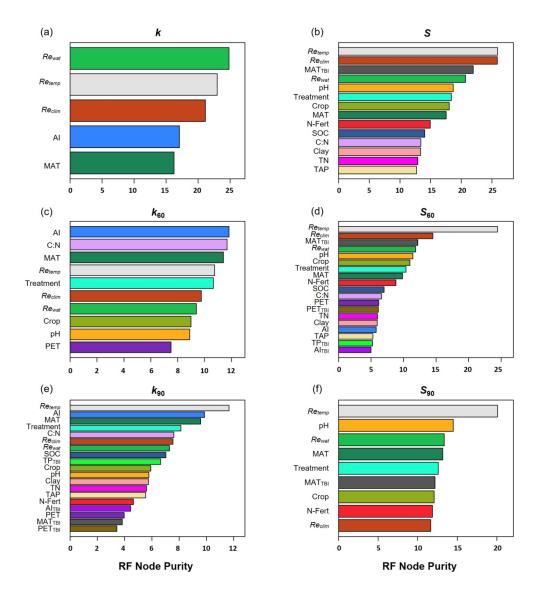






Figure caption 855 856 Figure 1 - Location and environmental zone of the Austrian and Swedish sites. 857 858 859 Figure 2 - Average decomposition rate (k) and stabilization (S) after the 90 days TBI period for each treatment and site in Austria. The extents of the box indicate 25th and 75th percentiles, and the 860 lines represent the 50th percentile. Whiskers represent the 10th and 90th percentiles and outliers are 861 862 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 863 864 2014. Sites AT2, AT3, AT5, AT7, AT9, and AT15 show results from 2015. And sites AT4, AT6, 865 AT8, AT10, AT11, AT12, AT13, AT14, and AT16 show results from 2016. 866 867 Figure 3 - Average decomposition rate (k) and stabilization (S) after the 90 days TBI period for each site and treatment in Sweden. The extents of the box indicate 25th and 75th percentiles, and 868 the lines represent the 50th percentile. Whiskers represent the 10th and 90th percentiles. 869 870 871 Figure 4 - Distribution of the mean decomposition rate constant (k) and the stabilization factor (S) 872 after the 90 days TBI period for each site in Austria and Sweden. Errors bars represent standard deviation. 873 874 Figure 5 - Variables selection procedure to identify the optimal number of variables to explain the 875 876 variance of k and S considering the combined dataset (60 and 90 days TBI period) with a Random Forest model. The blue point represents the optimal model. a) and b) Variables affecting k and S 877

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over all sampling times; c) and d) Variables affecting *k* and *S* after 60 days; e) and f) Variables affecting *k* and *S* after 90 days.

Figure 6 - Relative importance of the variables used by each optimized Random Forest model to predict the variance in the *k* and *S* parameters for the combined dataset (60 and 90 days TBI period) in Austria and Sweden. The higher the Node purity, the higher the importance of such variable. a) and b) Variables affecting *k* and *S* in all times; c) and d) Variables affecting *k* and *S* after 60 days; e) and f) Variables affecting *k* and *S* after 90 days.